abcam

ab247655

Anti-CD74 antibody [EPR4064] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR4064
Purification technique	Affinity purification Protein A
Concentration	1.046 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Not recommended

Species	Human, Synthetic peptide
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	Perform antigen retrieval before commencing with IHC staining protocol.

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

I-ELISA

Tested

Species	Synthetic peptide
Dilution info	-
Notes	-

Expected

Species	Human
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information CD74

Function	Plays a critical role in MHC class II antigen processing by stabilizing peptide-free class II alpha/beta heterodimers in a complex soon after their synthesis and directing transport of the complex from the endoplasmic reticulum to the endosomal/lysosomal system where the antigen processing and binding of antigenic peptides to MHC class II takes place. Serves as cell surface receptor for the cytokine MIF. Class-II-associated invariant chain peptide. Binds to the peptide-binding site of MHC class II alpha/beta heterodimers forming an alpha-beta-CLIP complex, thereby preventing the loading of antigenic peptides to the MHC class II complex until its release by HLA-DM in the endosome. Isoform p41. Stabilizes the conformation of mature CTSL by binding to its active site and serving as a chaperone to help maintain a pool of mature enzyme in endocytic compartments and extracellular space of antigen-presenting cells (APCs). Has antiviral activity by stymieing the endosomal entry of Ebola virus and coronaviruses, including SARS-CoV-2 (PubMed:32855215). Disrupts cathepsin-mediated Ebola virus glycoprotein processing, which prevents viral fusion and entry. This antiviral activity is specific to p41 isoform (PubMed:32855215).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab247655 is the carrier-free version of ab108393.

Species reactivity

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

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.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Supplementary info

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

Activity summary	CD74 also known as the invariant chain or Ii is a protein with a mass of about 33 kDa. It is widely expressed on the surface of immune cells such as B cells dendritic cells and macrophages. CD74 plays an important role in the immune system by acting as a chaperone for major histocompatibility complex class II (MHC II) molecules guiding their migration to endosomes where they encounter antigenic peptides. The CD74 protein also functions as a cell surface receptor for macrophage migration inhibitory factor (MIF) enhancing the immune response process.
Biological function summary	CD74 facilitates multiple immune regulatory processes. It forms a complex with the MHC II molecules aiding in their proper folding and peptide loading in the endoplasmic reticulum. This complex then travels through the Golgi apparatus towards the lysosomal compartments where antigens are presented to initiate adaptive immune responses. In addition CD74 is involved in signal transduction pathways that regulate cell proliferation and survival influenced by interactions with MIF and other molecules.
Pathways	The role of CD74 extends into both the antigen presentation and MIF signaling pathways. Within the antigen presentation pathway CD74's activity is tightly connected with MHC II and subsequently to CD4+ T cells facilitating the activation of these immune cells. The MIF signaling pathway involves CD74 interacting with CD44 a receptor that further propagates the signaling cascades essential for immune modulation and cell survival.
Associated diseases and disorders	CD74 is linked to inflammatory and autoimmune conditions such as rheumatoid arthritis and systemic lupus erythematosus. Aberrant expression of CD74 influences the presentation of autoantigens and may contribute to the chronic inflammation seen in these disorders. Its connection to MIF which is a pro-inflammatory cytokine further implicates CD74 in these autoimmune scenarios by promoting persistent immune activation. Researchers are also exploring CD74 as a therapeutic target in certain cancers due to its influence on immune surveillance and tumor progression.

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.

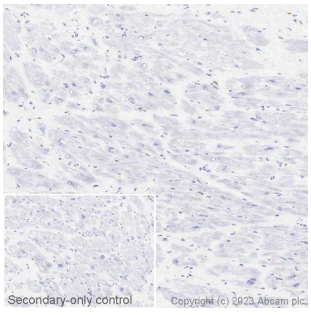
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

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15 product images



Immunohistochemistry (Frozen sections) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

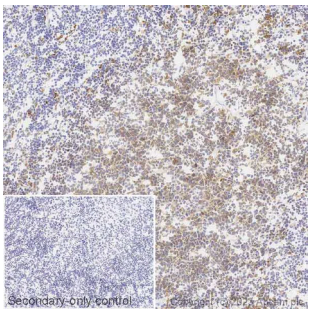
CD74 Immunohistochemistry (Frozen sections) staining of Human normal heart using rabbit Anti-CD74 antibody

Negative control image: IHC image of CD74 staining in a section of frozen human normal heart☒ performed on a Leica Biosystems BOND® RX instrument using the standard protocol. The section was fixed in 10% paraformaldehyde (10 min) prior to staining. The section was incubated with ab108393, 0.05ugml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset secondary-only control image is taken from an identical assay without primary antibody.

☒ Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA and sodium azide (ab108393).



Immunohistochemistry (Frozen sections) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

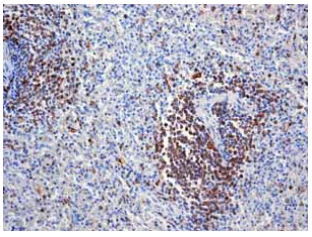
CD74 Immunohistochemistry (Frozen sections) staining of Human normal tonsil using rabbit Anti-CD74 antibody

IHC image of CD74 staining in a section of frozen human normal tonsil⊠ performed on a Leica Biosystems BOND® RX instrument using the standard protocol. The section was fixed in 10% paraformaldehyde (10 min) prior to staining. The section was incubated with ab108393, 0.05ugml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset secondary-only control image is taken from an identical assay without primary antibody.

⊠Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA and sodium azide (ab108393).

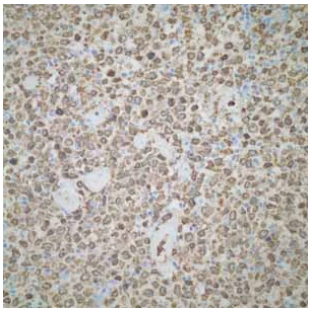


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CD74 antibody

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
ab108393 showing positive staining in Normal spleen tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

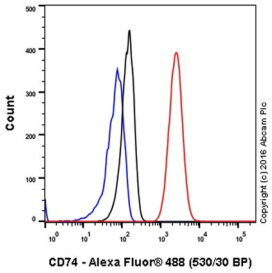


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CD74 antibody

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
ab108393 showing positive staining in B cell lymphoma tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

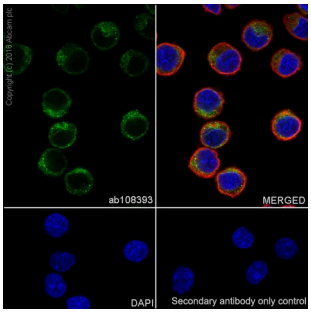


Flow Cytometry (Intracellular) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Flow Cytometry (Intracellular) staining using rabbit Anti-CD74 antibody

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
ab108393 staining CD74 in RAMOS (human Burkitt's lymphoma) cell line. The sample was fixed with 4% paraformaldehyde and incubated with the primary antibody (1/150) for 30 minutes at 4°C. An Alexa Fluor® 488 -conjugated goat anti-rabbit IgG (1/2000) was used as the secondary antibody.

Control: Cell without incubation with primary antibody and secondary antibody (Blue).

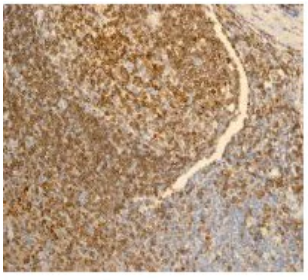


Immunocytochemistry/ Immunofluorescence - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CD74 antibody

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
Confocal image of ab108393 staining CD74 in the RAMOS (human Burkitt's lymphoma) cell line by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100 in PBS. Samples were incubated with primary antibody (1/500) and an Alexa Fluor® 488-conjugated goat anti-rabbit IgG polyclonal (ab150077) was used as the secondary antibody at a dilution of 1/1000.

DAPI was used ad a nuclear counterstain at 1/200.

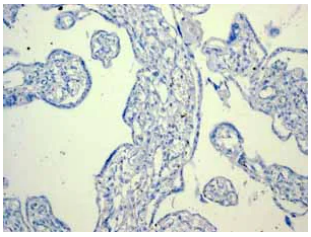


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CD74 antibody

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of CD74 in paraffin embedded Human tonsil tissue, using ab108393 at a 1/100 dilution.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

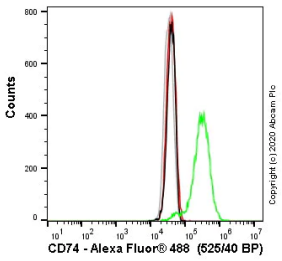


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CD74 antibody

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
ab108393 showing negative staining in Normal placenta tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



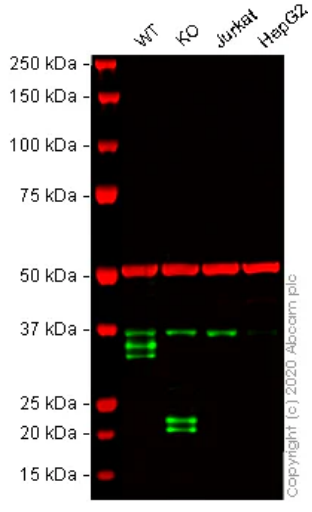
Flow Cytometry (Intracellular) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Flow Cytometry (Intracellular) staining using rabbit Anti-CD74 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab108393)
Intracellular Flow Cytometry overlay histogram showing wild-type Raji (green line) and CD74 knockout Raji cells (ab273378) stained with ab108393 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10µg/ml human IgG and 10% normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody (ab108393) ($1x10^6$ in 100µl at 0.2 µg/ml) for 30 min at 22°C.

The secondary antibody Goat anti-rabbit IgG H&L (Alexa Fluor® 488 pre-adsorbed) (ab150081) was used at 1/2000 for 30 min at 22°C. Isotype control antibody was Rabbit IgG (monoclonal) (ab172730) used at the same concentration and conditions as the primary antibody (wild-type Raji cells - black line; CD74 knockout Raji cells ab273378 - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity).

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.



Western blot - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Western blot staining using rabbit Anti-CD74 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab108393).
Lanes 1 - 4: Merged signal (red and green). Green - ab108393 observed at 35 kDa. Red - loading control, ab7291 (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.

ab108393 was shown to react with CD74 in western blot. The band observed in CD74 CRISPR/Cas9 edited cell line ab273378 (CRISPR/Cas9 edited lysate ab275529) below 35 kDa is likely to represent a truncated form. This has not been investigated further. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with ab108393 and ab7291 (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed

(ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.
All lanes:
Western blot - Anti-CD74 antibody [EPR4064] (ab108393) at 1/1000 dilution

Lane 1:
Wild-type Raji cell lysate at 30 µg

Lane 2:
Western blot - Human CD74 knockout Raji cell lysate (ab275529) at 30 µg

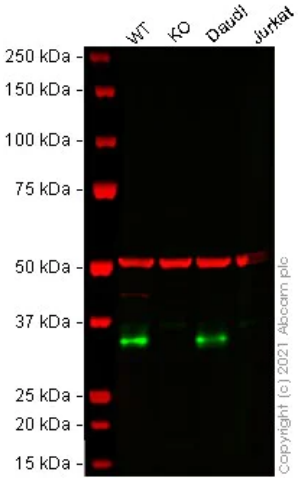
Lane 3:
Jurkat cell lysate at 30 µg

Lane 4:
HepG2 cell lysate at 30 µg

Performed under reducing conditions.

Predicted band size: 34 kDa

Observed band size: 35 kDa



Western blot - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Western blot staining using rabbit Anti-CD74 antibody

Lanes 1 - 4: Merged signal (red and green). Green - ab108393 observed at 34 kDa. Red - loading control ab7291 (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.
ab108393 was shown to react with CD74 in wild-type Raji cells in Western blot with loss of signal observed in CD74 knockout cell line ab273876 (knockout cell lysate ab273830). Wild-type Raji and CD74 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with ab108393 and ab7291 (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.

All lanes:
Western blot - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655) at 1/1000 dilution

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

Lane 2:
CD74 knockout Raji cell lysate at 20 µg

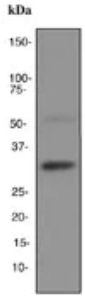
Lane 3:
Daudi cell lysate at 20 µg

Lane 4:
Jurkat cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 34 kDa

Observed band size: 34 kDa

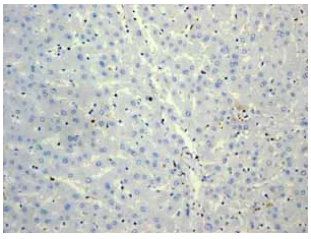


Western blot - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
All lanes:
Western blot - Anti-CD74 antibody [EPR4064] (ab108393) at 1/1000 dilution

All lanes:
Ramos cell lysate at 10 µg

Predicted band size: 34 kDa

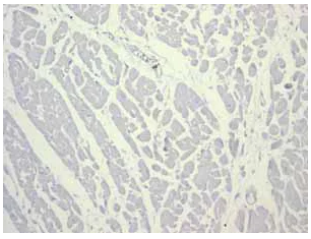


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CD74 antibody

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
ab108393 showing negative staining in Normal liver tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

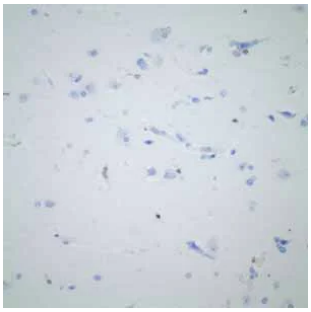


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CD74 antibody

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
ab108393 showing negative staining in Normal heart tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD74 antibody [EPR4064] - BSA and Azide free (ab247655)

CD74 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CD74 antibody

This data was developed using ab108393, the same antibody clone in a different buffer formulation.
ab108393 showing negative staining in Normal brain tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

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