abcam

ab225636

Anti-HLA Class 1 ABC antibody [EPR22172]

Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636) is a rabbit monoclonal antibody detecting HLA Class 1 ABC in **Western Blot, Flow Cytometry, IP, IHC-P, IHC-Fr, ICC/IF**. Suitable for **Human**.

- Biophysical QC for unrivalled batch-batch consistency

Recombinant | RabMAb | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR22172
Purification technique	Affinity purification Protein A
Concentration	0.659 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-Fr

Tested

Species	Human
Dilution info	0.05 µg/mL

Notes	-

Not recommended

Species	Recombinant fragment - Human
Dilution info	-
Notes	-

IP

Tested

Species	Human
Dilution info	1/30
Notes	-

Not recommended

Species	Recombinant fragment - Human
Dilution info	-
Notes	-

Flow Cyt

Tested

Species	Human
Dilution info	1/600
Notes	-

Not recommended

Species	Recombinant fragment - Human
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/1000
Notes	-

Species	Recombinant fragment - Human
Dilution info	1/1000
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/250
Notes	-

Not recommended

Species	Recombinant fragment - Human
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/4000
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Not recommended

Species	Recombinant fragment - Human

Dilution info	-
Notes	-

Target data

See full target information HLA-B

Function

Antigen-presenting major histocompatibility complex class I (MHCI) molecule. In complex with B2M/beta 2 microglobulin displays primarily viral and tumor-derived peptides on antigen-presenting cells for recognition by alpha-beta T cell receptor (TCR) on HLA-B-restricted CD8-positive T cells, guiding antigen-specific T cell immune response to eliminate infected or transformed cells (PubMed:23209413, PubMed:25808313, PubMed:29531227, PubMed:9620674). May also present self-peptides derived from the signal sequence of secreted or membrane proteins, although T cells specific for these peptides are usually inactivated to prevent autoreactivity (PubMed:18991276, PubMed:7743181). Both the peptide and the MHC molecule are recognized by TCR, the peptide is responsible for the fine specificity of antigen recognition and MHC residues account for the MHC restriction of T cells (PubMed:24600035, PubMed:29531227, PubMed:9620674). Typically presents intracellular peptide antigens of 8 to 13 amino acids that arise from cytosolic proteolysis via constitutive proteasome and IFNG-induced immunoproteasome (PubMed:23209413). Can bind different peptides containing allele-specific binding motifs, which are mainly defined by anchor residues at position 2 and 9 (PubMed:25808313, PubMed:29531227). Allele B*07:02: Displays peptides sharing a common signature motif, namely a Pro residue at position 2 and mainly a Leu anchor residue at the C-terminus (PubMed:7743181). Presents a long peptide (APRGPHGGAASGL) derived from the cancer-testis antigen CTAG1A/NY-ESO-1, eliciting a polyclonal CD8-positive T cell response against tumor cells (PubMed:29531227). Presents viral epitopes derived from HIV-1 gag-pol (TPQDLNTML) and Nef (RPQVPLRPM) (PubMed:25808313). Presents an immunodominant epitope derived from SARS-CoV-2 N/nucleoprotein (SPRWYFYYL) (PubMed:32887977). Displays self-peptides including a peptide derived from the signal sequence of HLA-DPB1 (APRTVALTA) (PubMed:7743181). Allele B*08:01: Presents to CD8-positive T cells viral epitopes derived from EBV/HHV-4 EBNA3 (QAKWRLQTL), eliciting cytotoxic T cell response. Allele B*13:02: Presents multiple HIV-1 epitopes derived from gag (RQANFLGKI, GQMREPRGSDI), nef (RQDILDLWI), gag-pol (RQYDQILIE, GQGQWTYQI) and rev (LQLPPLERL), all having in common a Gln residue at position 2 and mainly hydrophobic amino acids Leu, Ile or Val at the C-terminus. Associated with successful control of HIV-1 infection. Allele B*18:01: Preferentially presents octameric and nonameric peptides sharing a common motif, namely a Glu at position 2 and Phe or Tyr anchor residues at the C-terminus (PubMed:14978097, PubMed:18991276, PubMed:23749632). Presents an EBV/HHV-4 epitope derived from BZLF1 (SELEIKRY) (PubMed:23749632). May present to CD8-positive T cells an antigenic peptide derived from MAGEA3 (MEVDPIGHLY), triggering an anti-tumor immune response (PubMed:12366779). May display a broad repertoire of self-peptides with a preference for peptides derived from RNA-binding proteins (PubMed:14978097). Allele B*27:05: Presents to CD8-positive T cells immunodominant viral epitopes derived from HCV POLG (ARMILMTHF), HIV-1 gag (KRWIILGLNK), IAV NP (SRYWAIRTR), SARS-CoV-2 N/nucleoprotein (QRNAPRITF), EBV/HHV-4 EBNA4 (HRCQAIRKK) and EBV/HHV-4 EBNA6 (RRIYDLIEL), conferring longterm protection against viral infection (PubMed:15113903,

PubMed:18385228, PubMed:19139562, PubMed:32887977, PubMed:9620674). Can present self-peptides derived from cytosolic and nuclear proteins. All peptides carry an Arg at position 2 (PubMed:1922338). The peptide-bound form interacts with NK cell inhibitory receptor KIR3DL1 and inhibits NK cell activation in a peptide-specific way, being particularly sensitive to the nature of the amino acid side chain at position 8 of the antigenic peptide (PubMed:15657948, PubMed:8879234). KIR3DL1 fails to recognize HLA-B*27:05 in complex with B2M and EBV/HHV-4 EBNA6 (RRIYDLIEL) peptide, which can lead to increased activation of NK cells during infection (PubMed:15657948). May present an altered repertoire of peptides in the absence of TAP1-TAP2 and TAPBPL (PubMed:9620674). Allele B*40:01: Presents immunodominant viral epitopes derived from EBV/HHV-4 LMP2 (IEDPPFNSL) and SARS-CoV-2 N/nucleoprotein (MEVTPSGTWL), triggering memory CD8-positive T cell response (PubMed:18991276, PubMed:32887977). Displays self-peptides sharing a signature motif, namely a Glu at position 2 and a Leu anchor residue at the C-terminus (PubMed:18991276). Allele B*41:01: Displays self-peptides sharing a signature motif, namely a Glu at position 2 and Ala or Pro anchor residues at the C-terminus. Allele B*44:02: Presents immunodominant viral epitopes derived from EBV/HHV-4 EBNA4 (VEITPYKPTW) and EBNA6 (AEGGVGWRHW, EENLLDFVRF), triggering memory CD8-positive T cell response (PubMed:18991276, PubMed:9620674). Displays self-peptides sharing a signature motif, namely a Glu at position 2 and Phe, Tyr or Trp anchor residues at the C-terminus (PubMed:18991276). Allele B*45:01: Displays self-peptides sharing a signature motif, namely a Glu at position 2 and Ala or Pro anchor residues at the C-terminus. Allele B*46:01: Preferentially presents nonameric peptides sharing a signature motif, namely Ala and Leu at position 2 and Tyr, Phe, Leu, or Met anchor residues at the C-terminus. The peptide-bound form interacts with KIR2DL3 and inhibits NK cell cytotoxic response in a peptide-specific way. Allele B*47:01: Displays self-peptides sharing a signature motif, namely an Asp at position 2 and Leu or Met anchor residues at the C-terminus. Allele B*49:01: Displays self-peptides sharing a signature motif, namely a Glu at position 2 and Ile or Val anchor residues at the C-terminus. Allele B*50:01: Displays self-peptides sharing a signature motif, namely a Glu at position 2 and Ala or Pro anchor residues at the C-terminus. Allele B*51:01: Presents an octameric HIV-1 epitope derived from gag-pol (TAFTIPSI) to the public TRAV17/TRBV7-3 TCR clonotype, strongly suppressing HIV-1 replication. Allele B*54:01: Displays peptides sharing a common signature motif, namely a Pro residue at position 2 and Ala anchor residue at the C-terminus. Allele B*55:01: Displays peptides sharing a common signature motif, namely a Pro residue at position 2 and Ala anchor residue at the C-terminus. Allele B*56:01: Displays peptides sharing a common signature motif, namely a Pro residue at position 2 and Ala anchor residue at the C-terminus. Allele B*57:01: The peptide-bound form recognizes KIR3DL1 and inhibits NK cell cytotoxic response. Presents HIV gag peptides (immunodominant KAFSPEVIPMF and subdominant KALGPAATL epitopes) predominantly to CD8-positive T cell clones expressing a TRAV41-containing TCR, triggering HLA-B-restricted T cell responses. Allele B*67:01: Displays peptides sharing a common signature motif, namely a Pro residue at position 2 and Leu anchor residue at the C-terminus.

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

What is this antibody validated in?
Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Flow Cyt), Immunoprecipitation (IP), Immunohistochemistry (IHC-P), Immunohistochemistry (IHC-Fr), Immunocytochemistry/immunofluorescence (ICC/IF) in Human samples.

What is the molecular weight of HLA Class 1 ABC?
Anti-HLA Class 1 ABC [EPR22172] (ab225636) specifically detects a band for HLA Class 1 ABC (UniProt: P01889) at a molecular weight of 40kDa.

Other related products
We have a range of other formats of antibody clone [EPR22172] also available for your convenience: ab225636, Carrier free - ab239788, Alexa Fluor® 555 - ab314336, Alexa Fluor® 488 - ab314926, Alexa Fluor® 647 - ab315134

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.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size 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.

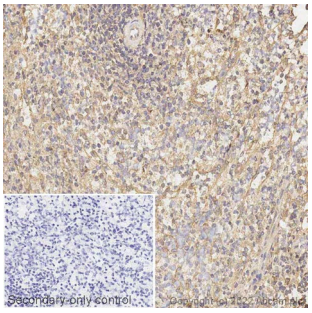
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

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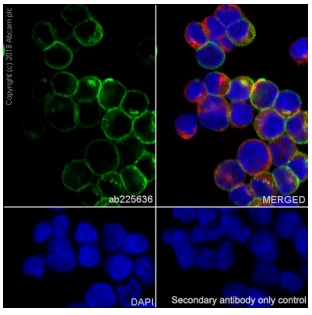
12 product images



Immunohistochemistry (Frozen sections) - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

IHC image of HLA Class 1 ABC staining in a section of human normal frozen spleen* 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 ab225636, 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. 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. *Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre

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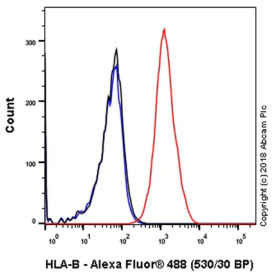


Immunocytochemistry/ Immunofluorescence - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

HLA Class 1 ABC Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-HLA Class 1 ABC antibody

Immunofluorescent analysis of 100% methanol-fixed Raji (human Burkitt's lymphoma cell line) cells labeling HLAB with ab225636 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing membranous staining in Raji cells. The nuclear counterstain is DAPI (blue).
Tubulin is detected with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa

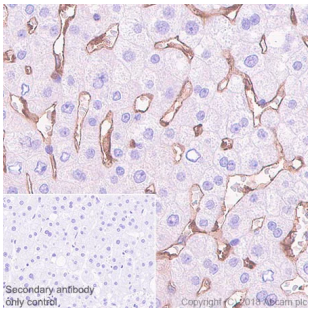
Fluor® 594) at 1/200 dilution (red).
The negative control is the secondary antibody only.
Methanol is recommended for fixation as weak signal was observed using 4% PFA.



Flow Cytometry - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

HLA Class 1 ABC Flow Cytometry staining using rabbit Anti-HLA Class 1 ABC antibody

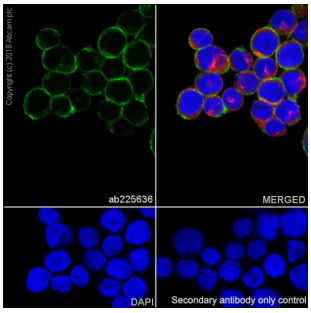
Flow cytometric analysis of Raji (human Burkitt's lymphoma cell line) cells labeling HLAB with ab225636 at 1/600 (red) compared with a Rabbit monoclonal IgG (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti-rabbit IgG (Alexa Fluor® 488, ab150077), at 1/2000 dilution was used as the secondary antibody.
Gated on viable cells.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

HLA Class 1 ABC Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-HLA Class 1 ABC antibody

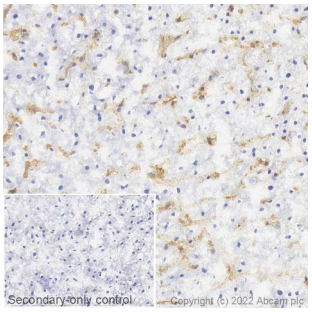
Immunohistochemical analysis of paraffin-embedded human liver tissue labeling HLAB with ab225636 at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining on hepatic sinus endothelium of human liver (PMID: 26963506) is observed. Counter stained with hematoxylin.
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).
Perform heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).



Immunocytochemistry/ Immunofluorescence - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

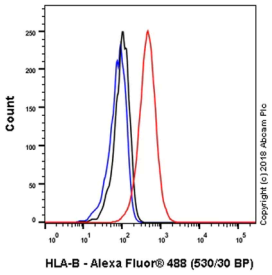
HLA Class 1 ABC Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-HLA Class 1 ABC antibody

Immunofluorescent analysis of 100% methanol-fixed THP-1 (human monocytic leukemia cell line) cells labeling HLAB with ab225636 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing membranous staining in THP-1 cells. The nuclear counterstain is DAPI (blue).
Tubulin is detected with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1/200 dilution (red).
The negative control is the secondary antibody only.
Methanol is recommended for fixation as weak signal was observed using 4% PFA.



Immunohistochemistry (Frozen sections) - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

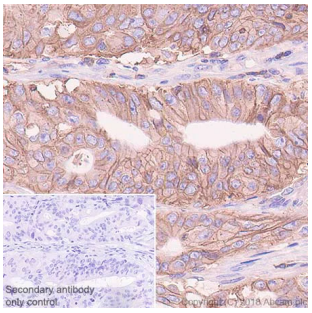
IHC image of HLA Class 1 ABC staining in a section of human normal frozen liver* performed on a Leica Biosystems BOND® RX instrument using the standard protocol. The section was fixed in 10% paraformaldehyde (10 min) prior to staining. The section was incubated with ab225636, 0.05ugml, for 15 mins at 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. 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. *Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Flow Cytometry - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

HLA Class 1 ABC Flow Cytometry staining using rabbit Anti-HLA Class 1 ABC antibody

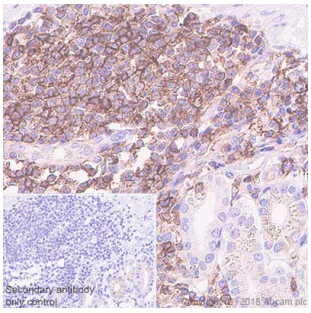
Flow cytometric analysis of THP-1 (human monocytic leukemia cell line) cell line labeling HLAB with ab225636 at 1/600 (red) compared with a Rabbit monoclonal IgG (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077), at 1/2000 dilution was used as the secondary antibody.
Gated on viable cells.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

HLA Class 1 ABC Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-HLA Class 1 ABC antibody

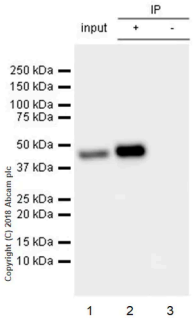
Immunohistochemical analysis of paraffin-embedded human colonic carcinoma tissue labeling HLAB with ab225636 at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Membranous and cytoplasmic staining on tumor cells of human colonic carcinoma (PMID: 26310568, PMID: 17014712) is observed. Counter stained with hematoxylin.
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).
Perform heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

HLA Class 1 ABC Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-HLA Class 1 ABC antibody

Immunohistochemical analysis of paraffin-embedded human stomach tissue labeling HLAB with ab225636 at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Membranous and cytoplasmic staining on lymph nodule of human stomach (PMID:25294906) is observed. Counter stained with hematoxylin.
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).
Perform heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).

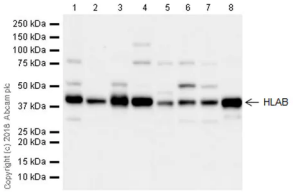


Immunoprecipitation - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

HLAB was immunoprecipitated from 0.35 mg THP-1 (human monocytic leukemia cell line) whole cell lysate with ab225636 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab225636 at 1/500 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1000 dilution.
Lane 1: THP-1 whole cell lysate 10 µg (Input).
Lane 2: ab225636 IP in THP-1 whole cell lysate.
Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab225636 in THP-1 whole cell lysate.
Blocking/Dilution buffer: 5% NFDM/TBST.
Exposure time: 3 seconds.
All lanes:
Immunoprecipitation - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

Predicted band size: 44 kDa

Observed band size: 40 kDa, 43 kDa



Western blot - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

HLA Class 1 ABC Western blot staining using rabbit Anti-HLA Class 1 ABC antibody

Blocking/Dilution buffer: 5% NFDM/TBST.
Weak reactive bands of undetermined proteins are observed in addition to the target band.

All lanes:
Western blot - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636) at 1/1000 dilution

Lane 1:
Raji (human Burkitt's lymphoma cell line), whole cell lysate at 20 µg

Lane 2:
THP-1 (human monocytic leukemia cell line), whole cell lysate at 20 µg

Lane 3:
HL-60 (human promyelocytic leukemia cell line), whole cell lysate at 20 µg

Lane 4:
Jurkat (human T cell leukemia cell line from peripheral blood), whole cell lysate at 20 µg

Lane 5:

HepG2 (human liver hepatocellular carcinoma cell line), whole cell lysate at 20 µg

Lane 6:
TF-1 (human bone marrow erythroleukemia cell line), whole cell lysate at 20 µg

Lane 7:
Ramos (human Burkitt's lymphoma cell line), whole cell lysate at 20 µg

Lane 8:
Human tonsil at 20 µg

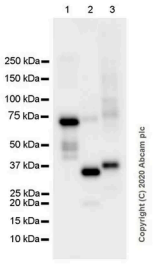
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/50000 dilution

Predicted band size: 40 kDa

Observed band size: 40 kDa

Exposure time: 26s



Western blot - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636)

HLA Class 1 ABC Western blot staining using rabbit Anti-HLA Class 1 ABC antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST.
This antibody has cross reaction with human HLA-A and HLA-C.

Recombinant human HLA-A protein was purchased from abnova company (H00003105-P01).

Recombinant human HLA-B protein was made in house and extracted from a E.coli expressoin system.

Recombinant human HLA-C protein was purchased from novusbio company (NBP2-23210).

All lanes:
Western blot - Anti-HLA Class 1 ABC antibody [EPR22172] (ab225636) at 1/1000 dilution

Lane 1:
Recombinant human HLA-A protein (aa24-aa365) at 10 ng

Lane 2:
Recombinant human HLA-B protein (aa25-aa299) at 10 ng

Lane 3:
Recombinant human HLA-C protein (aa25-aa308) at 10 ng

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 63 kDa, 34 kDa, 37 kDa

Exposure time: 3s

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