# abcam

## Product datasheet

# Anti-HLA E antibody [MEM-E/02] ab2216



★★★★ 6 Abreviews 18 References 5 Images

Overview

Product name Anti-HLA E antibody [MEM-E/02]

**Description** Mouse monoclonal [MEM-E/02] to HLA E

Host species Mouse

Specificity This antibody reacts with the denaturated heavy chain of human HLA-E. It does not cross-react

with HLA-A, -B, -C or -G. Specifity of the antibody was confirmed on HLA-G/HLA-E

Workshop(Victoria 2002).

Tested applications Suitable for: Flow Cyt, WB, IHC-P

Species reactivity Reacts with: Human

**Immunogen** Recombinant full length protein corresponding to Human HLA E.

Database link: P13747

Positive control WB: A549, THP-1 and Jurkat cell lysates. IHC-P: Human tonsil tissue. Flow Cyt: HL60 cells.

**General notes**This product has been changed from ascites to tissue culture supernatant. Please note that the

dilutions may need to be adjusted accordingly. If you have any questions, please do not hesitate to

contact our scientific support team.

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** pH: 7.40

Preservative: 0.097% Sodium azide

Constituent: PBS

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**Purity** Protein A purified

**Purification notes** Purified from TCS. Purity >95% by SDS-PAGE.

Clonality Monoclonal
Clone number MEM-E/02
Myeloma unknown
Isotype IgG1

**Light chain type** unknown

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab2216 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
Flow Cyt		Use 1µg for 10 <sup>6</sup> cells.  ab170190 - Mouse monoclonal lgG1, is suitable for use as an isotype control with this antibody.
WB	<b>★★★★ (4)</b>	Use at an assay dependent concentration. Predicted molecular weight: 40 kDa.
IHC-P	★★☆☆☆ (1)	Use a concentration of 5 - 10 μg/ml.

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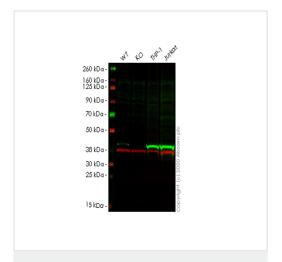
Relevance HLA E belongs to the HLA class I heavy chain paralogues. This class I molecule is a heterodimer

consisting of a heavy chain and a light chain (beta-2 microglobulin). The heavy chain is anchored in the membrane. HLA E binds a restricted subset of peptides derived from the leader peptides

of other class I molecules.

Cellular localization Membrane; Single-pass type I membrane protein

# **Images**



Western blot - Anti-HLA E antibody [MEM-E/02] (ab2216)

**All lanes :** Anti-HLA E antibody [MEM-E/02] (ab2216) at 1/500 dilution

Lane 1: Wild-type A549 cell lysate

Lane 2: HLA-E knockout A549 cell lysate

Lane 3 : THP-1 cell lysate

Lane 4 : Jurkat cell lysate

Lysates/proteins at 20 µg per lane.

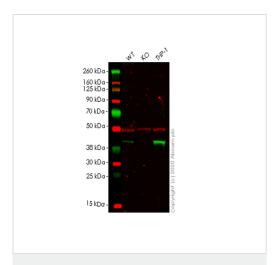
#### Secondary

**All lanes :** Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (ab216777) at 1/10000 dilution

Predicted band size: 40 kDa Observed band size: 40 kDa

**Lanes 1-4:** Merged signal (red and green). Green - ab2216 observed at 40 kDa. Red - loading control **ab181602** observed at 36 kDa.

ab2216 Anti-HLA E antibody [MEM-E/02] was shown to specifically react with HLA E in wild-type A549 cells. Loss of signal was observed when knockout cell line <a href="mailto:ab267080">ab267080</a> (knockout cell lysate <a href="mailto:ab258452">ab258452</a>) was used. Wild-type and HLA E knockout samples were subjected to SDS-PAGE. ab2216 and Anti-GAPDH antibody[EPR16891] - Loading Control (<a href="mailto:ab181602">ab181602</a>) were incubated at room temperature for 2.5 hours at 1 in 500 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 680RD) preadsorbed (<a href="mailto:ab216777">ab216777</a>) and Goat anti-Mouse lgG H&L (IRDye® 800CW) preadsorbed (<a href="mailto:ab216772">ab216777</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-HLA E antibody [MEM-E/02] (ab2216)

**All lanes :** Anti-HLA E antibody [MEM-E/02] (ab2216) at 1/500 dilution

Lane 1: Wild-type HEK-293T cell lysate

Lane 2: HLA-E knockout HEK-293T cell lysate

Lane 3: THP-1 cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

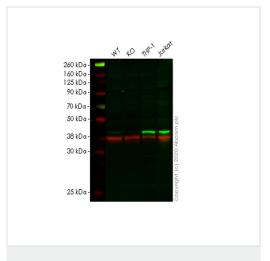
**All lanes :** Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (<u>ab216777</u>) at 1/10000 dilution

Performed under reducing conditions.

Predicted band size: 40 kDa

**Lanes 1-3:** Merged signal (red and green). Green - ab2216 observed at 40 kDa. Red - loading control **ab52901** observed at kDa.

ab2216 Anti-HLA E antibody [MEM-E/02] was shown to specifically react with HLA E in wild-type HEK293T cells. Loss of signal was observed when knockout cell line <a href="mailto:ab267231">ab267231</a> (knockout cell lysate <a href="mailto:ab258454">ab258454</a>) was used. Wild-type and HLA E knockout samples were subjected to SDS-PAGE. ab2216 and Anti-beta Tubulin [EP1331Y] - Microtubule Marker (<a href="mailto:ab52901">ab52901</a>) were incubated overnight at 4°C at 1 in 500 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 680RD) preadsorbed (<a href="mailto:ab216777">ab216777</a>) and Goat anti-Mouse lgG H&L (IRDye® 800CW) preadsorbed (<a href="mailto:ab216772">ab216772</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-HLA E antibody [MEM-E/02] (ab2216)

**All lanes :** Anti-HLA E antibody [MEM-E/02] (ab2216) at 1/1000 dilution

Lane 1: Wild-type A549 cell lysate

Lane 2: HLA-E knockout A549 cell lysate

Lane 3 : THP-1 cell lysate

Lane 4 : Jurkat cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (<u>ab216777</u>) at 1/10000 dilution

Predicted band size: 40 kDa

**Lanes 1-4:** Merged signal (red and green). Green - ab2216 observed at 40 kDa. Red - loading control <u>ab181602</u> observed at 36 kDa.

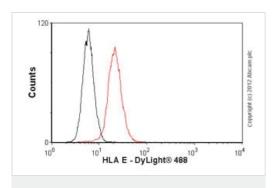
ab2216 Anti-HLA E antibody [MEM-E/02] was shown to specifically react with HLA E in wild-type A549 cells. Loss of signal was observed when knockout cell line <a href="mailto:ab267081">ab267081</a> (knockout cell lysate <a href="mailto:ab258453">ab258453</a>) was used. Wild-type and HLA E knockout samples were subjected to SDS-PAGE. ab2216 and Anti-GAPDH antibody[EPR16891] - Loading Control (<a href="mailto:ab181602">ab181602</a>) were incubated at room temperature for 2.5 hours at 1 in 500 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 680RD) preadsorbed (<a href="mailto:ab216777">ab216777</a>) and Goat anti-Mouse lgG H&L (IRDye® 800CW) preadsorbed (<a href="mailto:ab216772">ab216777</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-HLA E antibody [MEM-E/02] (ab2216)

IHC image of ab2216 staining in human tonsil formalin fixed paraffin embedded tissue section, performed on a Leica Bond<sup>TM</sup> system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab2216, 1µg/ml, 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.

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.



Flow Cytometry - Anti-HLA E antibody [MEM-E/02] (ab2216)

Overlay histogram showing HL60 cells stained with ab2216 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. 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 (ab2216, 1 µg/1x10<sup>6</sup> cells for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was Mouse IgG1 [ICIGG1] (ab91353, 2 µg/1x10<sup>6</sup> cells) used under the same conditions. Acquisition of >5,000 events was performed.

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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