

Human HLA-E (HLA E) knockout A549 cell line ab267080

画像数 3

製品の概要

製品名	Human HLA-E (HLA E) knockout A549 cell line
Parental Cell Line	A549
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 1 bp insertion in exon 2
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
アプリケーション	適用あり: WB
Biosafety level	2
特記事項	<p>Recommended control: Human wild-type A549 cell line (ab255450). Please note a wild-type cell line is not automatically included with a knockout cell line order, if required please add recommended wild-type cell line at no additional cost using the code WILDTYPE-TMTK1.</p> <p>Cryopreservation cell medium: Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose.</p> <p>Culture medium: F-12K + 10% FBS</p> <p>Initial handling guidelines: Upon arrival, the vial should be stored in liquid nitrogen vapor phase and not at -80°C. Storage at -80°C may result in loss of viability.</p> <ol style="list-style-type: none"> 1. Thaw the vial in 37°C water bath for approximately 1-2 minutes. 2. Transfer the cell suspension (0.8 mL) to a 15 mL/50 mL conical sterile polypropylene centrifuge tube containing 8.4 mL pre-warmed culture medium, wash vial with an additional 0.8 mL culture medium (total volume 10 mL) to collect remaining cells, and centrifuge at 201 x g (rcf) for 5 minutes at room temperature. 10 mL represents minimum recommended dilution. 20 mL represents maximum recommended dilution. 3. Resuspend the cell pellet in 5 mL pre-warmed culture medium and count using a haemocytometer or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of 2×10^3-1×10^4 cells/cm². Seeding density is given as a guide only and should be scaled to align with individual lab schedules. 4. Incubate the culture at 37°C incubator with 5% CO₂. Cultures should be monitored daily. <p>Subculture guidelines:</p> <p>All seeding densities should be based on cell counts gained by established methods. A guide seeding density of 6×10^4 cells/cm² is recommended.</p> <p>A partial media change 24 hours prior to subculture may be helpful to encourage growth, if required.</p>

Cells should be passaged when they have achieved 80-90% confluence.

Do not exceed 7×10^4 cells/cm².

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We will provide viable cells that proliferate on revival.

製品の特性

Number of cells	1 x 10 ⁶ cells/vial, 1 mL
Adherent /Suspension	Adherent
Tissue	Lung
Cell type	epithelial
Disease	Carcinoma
Gender	Male
STR Analysis	Amelogenin X,YD5S818: 11 D13S317: 11 D7S820: 8, 11 D16S539: 11, 12 vWA: 14 TH01: 8,9.3 TPOX: 8,11 CSF1PO: 10, 12
Mycoplasma free	Yes
保存方法	Shipped on Dry Ice. Store in liquid nitrogen.
バッファー	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

ターゲット情報

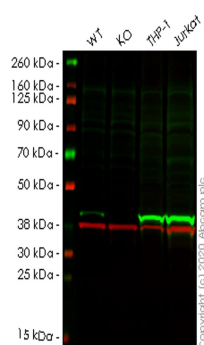
関連性	HLA E belongs to the HLA class I heavy chain paralogues. This class I molecule is a heterodimer consisting of a heavy chain and a light chain (beta-2 microglobulin). The heavy chain is anchored in the membrane. HLA E binds a restricted subset of peptides derived from the leader peptides of other class I molecules.
細胞内局在	Membrane; Single-pass type I membrane protein

アプリケーション

The Abpromise guarantee **Abpromise保証は、**次のテスト済みアプリケーションにおけるab267080の使用に適用されます
アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration. Predicted molecular weight: 40 kDa.

画像



Western blot - Human HLA-E (HLA E) knockout A549 cell line (ab267080)

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.

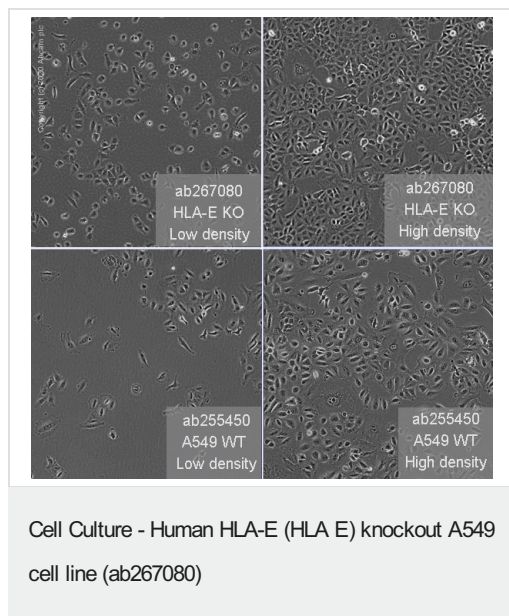
ab2216 Anti-HLA E antibody [MEM-E/02] was shown to specifically react with HLA E in wild-type A549 cells. Loss of signal was observed when knockout cell line ab267080 (knockout cell lysate **ab258452**) was used. Wild-type and HLA E knockout samples were subjected to SDS-PAGE. **ab2216** and Anti-GAPDH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

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Mut  AGGGGTGAGAGTATTGGGACCGGGAGACACGGAGCCGCCAGGGACCCGACAGATTTTC
      |||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
WT   AGGGGTGAGAGTATTGGGACCGGGAGACACGGAGC GCCAGGGACCCGACAGATTTTC
  
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Sanger Sequencing - Human HLA-E knockout A549 cell line (ab267080)

Homozygous: 1 bp insertion in exon2



Representative images of HLA-E knockout A549 cells, low and high confluency examples (top left and right respectively) and wild-type A549 cells, low and high confluency (bottom left and right respectively) showing typical adherent, epithelial-like morphology. Images were captured at 10X magnification using an EVOS M5000 microscope.

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