

Human L1CAM knockout HeLa cell line ab255401

3 Images

Overview

Product name	Human L1CAM knockout HeLa cell line
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 154 bp insertion in exon 1
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
Tested applications	Suitable for: WB
Biosafety level	2
General notes	<p>Recommended control: Human wild-type HeLa cell line (ab255928). 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: DMEM (High Glucose) + 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 2×10^4 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 2×10^4 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.

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We will provide viable cells that proliferate on revival.

Properties

Number of cells	1 x 10 ⁶ cells/vial, 1 mL
Adherent /Suspension	Adherent
Tissue	Cervix
Cell type	epithelial
Disease	Adenocarcinoma
Gender	Female
STR Analysis	Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 WWA: 16, 18 TH01: 7 TPOX: 8,12 CSF1PO: 9, 10
Mycoplasma free	Yes
Storage instructions	Shipped on Dry Ice. Store in liquid nitrogen.
Storage buffer	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

Target

Function	Cell adhesion molecule with an important role in the development of the nervous system. Involved in neuron-neuron adhesion, neurite fasciculation, outgrowth of neurites, etc. Binds to axonin on neurons.
Involvement in disease	<p>Defects in L1CAM are the cause of hydrocephalus due to stenosis of the aqueduct of Sylvius (HSAS) [MIM:307000]. Hydrocephalus is a condition in which abnormal accumulation of cerebrospinal fluid in the brain causes increased intracranial pressure inside the skull. This is usually due to blockage of cerebrospinal fluid outflow in the brain ventricles or in the subarachnoid space at the base of the brain. In children is typically characterized by enlargement of the head, prominence of the forehead, brain atrophy, mental deterioration, and convulsions. In adults the syndrome includes incontinence, imbalance, and dementia. HSAS is characterized by mental retardation and enlarged brain ventricles.</p> <p>Defects in L1CAM are the cause of mental retardation-aphasia-shuffling gait-adducted thumbs syndrome (MASA) [MIM:303350]; also known as corpus callosum hypoplasia, psychomotor retardation, adducted thumbs, spastic paraparesis, and hydrocephalus or CRASH syndrome. MASA is an X-linked recessive syndrome with a highly variable clinical spectrum. Main clinical features include spasticity and hyperreflexia of lower limbs, shuffling gait, mental retardation, aphasia and adducted thumbs. The features of spasticity have been referred to as complicated spastic paraplegia type 1 (SPG1). Some patients manifest corpus callosum hypoplasia and hydrocephalus. Inter- and intrafamilial variability is very wide, such that patients with hydrocephalus, MASA, SPG1, and agenesis of corpus callosum can be present within the same family.</p> <p>Defects in L1CAM are the cause of spastic paraplegia X-linked type 1 (SPG1) [MIM:303350]. Spastic paraplegia is a degenerative spinal cord disorder characterized by a slow, gradual,</p>

progressive weakness and spasticity of the lower limbs.

Note=Defects in L1CAM may contribute to Hirschsprung disease by modifying the effects of Hirschsprung disease-associated genes to cause intestinal aganglionosis.

Defects in L1CAM are a cause of partial agenesis of the corpus callosum (ACCPX)

[MIM:304100]. A syndrome characterized by partial corpus callosum agenesis, hypoplasia of inferior vermis and cerebellum, mental retardation, seizures and spasticity. Other features include microcephaly, unusual facies, and Hirschsprung disease in some patients.

Sequence similarities

Belongs to the immunoglobulin superfamily. L1/neurofascin/NgCAM family.

Contains 5 fibronectin type-III domains.

Contains 6 Ig-like C2-type (immunoglobulin-like) domains.

Cellular localization

Cell membrane.

Applications

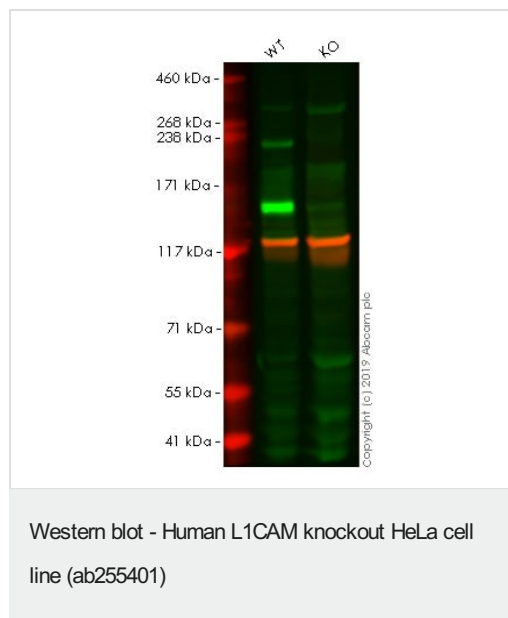
The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab255401 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
WB		Use at an assay dependent concentration. Predicted molecular weight: 140 kDa.

Images



All lanes : Anti-L1CAM antibody [EPR18750] (**ab208155**) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate

Lane 2 : L1CAM knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/20000 dilution

Performed under reducing conditions.

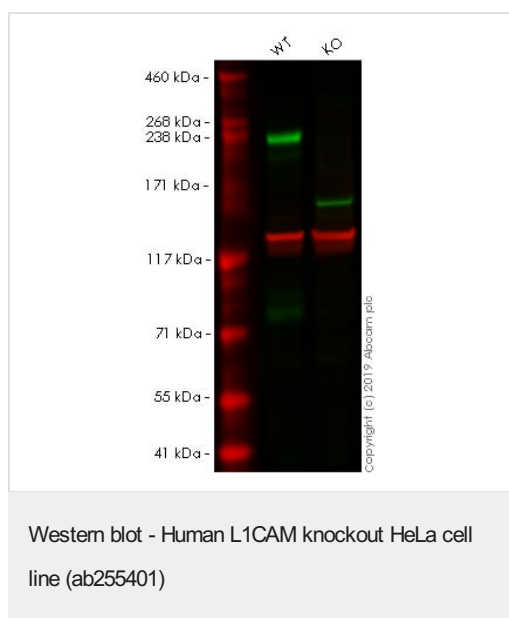
Predicted band size: 140 kDa

Additional bands at: 124 kDa (possible Loading Control)

Lanes 1 -2: Merged signal (red and green). Green - **ab208155** observed at 220 kDa. Red - loading control, **ab130007** observed at

125 kDa.

ab208155 was shown to react with L1CAM in wild-type HeLa. Loss of signal was observed when knockout cell line ab255401 (knockout cell lysate **ab263786**) was used. Wild-type and L1CAM knockout samples were subjected to SDS-PAGE. **ab208155** and Anti-Vinculin antibody [VIN-54] (**ab130007**) were incubated overnight at 4°C 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 : Anti-L1CAM antibody [EPR18998] (**ab182407**) at 1/5000 dilution

Lane 1 : Wild-type HeLa cell lysate

Lane 2 : L1CAM knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/20000 dilution

Performed under reducing conditions.

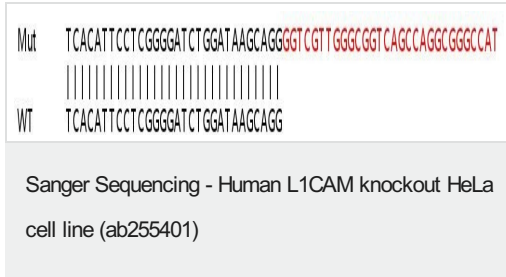
Predicted band size: 140 kDa

Additional bands at: 124 kDa (possible Loading Control)

Lanes 1 - 2: Merged signal (red and green). Green - **ab182407** observed at 220 kDa. Red - loading control, **ab130007** observed at 125 kDa.

ab182407 was shown to react with L1CAM in wild-type HeLa. Loss of signal was observed when knockout cell line ab255401 (knockout cell lysate **ab263786**) was used. Wild-type and L1CAM knockout samples were subjected to SDS-PAGE. **ab182407** and Anti-Vinculin antibody [VIN-54] (**ab130007**) were incubated overnight at 4°C at 1 in 5000 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.



Homozygous: 154 bp insertion in exon1

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