abcam

Product datasheet

Human CXCL10 (IP10) knockout THP-1 cell line ab277860

3 Images

Overview

Product name	Human CXCL10 (IP10) knockout THP-1 cell line
Parental Cell Line	THP-1
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 80 bp Deletion in Exon 2
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
Tested applications	**Suitable for:** WB
Biosafety level	1
General notes	**Recommended control:** Human wild-type THP-1 cell line (**ab281894**). 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. **Cryopreservation cell medium:** Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose. **Culture medium:** RPMI + 10% FBS + 0.05 mM beta-mercaptoethanol **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. 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 2-4x10^5 cells/mL. 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% CO_2. Cultures should be monitored daily. 5. THP-1 cells recover slowly from cryopreservation and therefore may not be ready for subculture for a number of days. Cells should be left as much as possible over this time and only subcultured when the cell density reaches 8x10^5 cells/mL.

Subculture guidelines:

All seeding densities should be based on cell counts gained by established methods.
Cells should be seeded at 2-4x10^5 cells/mL and subcultured when they have reached 8x10^5 cells/mL. It is not recommended to allow the cell density to exceed 1x10^6 cells/mL.
A partial media change 24 hours prior to subculture may be helpful to encourage growth, if required.
Cells should be passaged when they have achieved 80-90% confluence.
This product is subject to limited use licenses from The Broad Institute and ERS Genomics Limited, and is developed with patented technology. For full details of the limited use licenses and relevant patents please refer to our **limited use license** and **patent pages**.

We will provide viable cells that proliferate on revival.

Properties

Number of cells	1 x 10^6 cells/vial, 1 mL
Adherent /Suspension	Suspension
Tissue	Blood
Cell type	acute monocytic leukemia
Disease	Acute Monocytic Leukemia
Gender	Male
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	Chemotactic for monocytes and T-lymphocytes. Binds to CXCR3.
Sequence similarities	Belongs to the intercrine alpha (chemokine CxC) family.
Post-translational modifications	CXCL10(1-73) is produced by proteolytic cleavage after secretion from keratinocytes.
Cellular localization	Secreted.

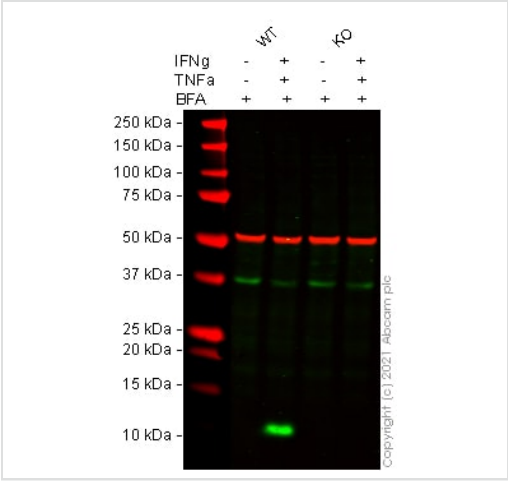
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab277860 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: 10 kDa.

Images



Western blot - Human CXCL10 (IP10) knockout THP-1 cell line (ab277860)

All lanes : Anti-IP10 antibody [EPR7850] (**ab137018**) at 1/1000 dilution

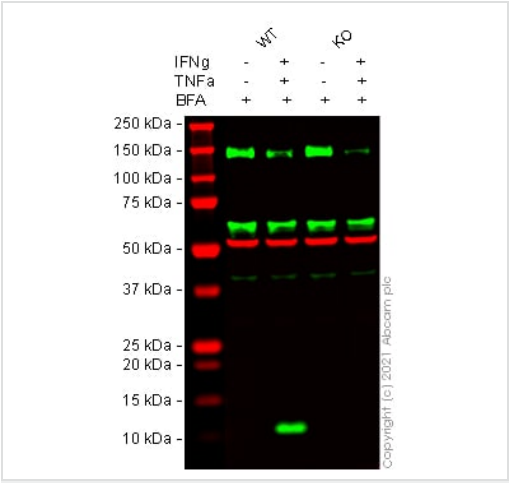
Lane 1 : Wild-type THP-1 vehicle control IFNg (0 ng/ml, 32 h), TNF-alpha (0 ng/ml, 32 h), Brefeldin A (5 ug/ml, 6 h) cell lysate
Lane 2 : Wild-type THP-1 treated IFNg (100 ng/ml, 32 h), TNF-alpha (10 ng/ml, 32 h), Brefeldin A (5 ug/ml, 6 h) cell lysate
Lane 3 : CXCL10 knockout THP-1 vehicle control IFNg (0 ng/ml, 32 h), TNF-alpha (0 ng/ml, 32 h), Brefeldin A (5 ug/ml, 6 h) cell lysate
Lane 4 : CXCL10 knockout THP-1 treated IFN-gamma (100 ng/ml, 32 h), TNF-alpha (10 ng/ml, 32 h), Brefeldin A (5 ug/ml, 6 h) cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 10 kDa
Observed band size: 11 kDa

False colour image of Western blot: Anti-IP10 antibody [EPR7850] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (**ab7291**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab137018** was shown to bind specifically to IP10. A band was observed at 11 kDa in treated wild-type THP-1 cell lysates with no signal observed at this size in treated CXCL10 knockout cell line ab277860 (knockout cell lysate **ab282997**). To generate this image, wild-type and CXCL10 knockout THP-1 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) at 1/20000 dilution.



Western blot - Human CXCL10 (IP10) knockout THP-1 cell line (ab277860)

All lanes : Anti-IP10 antibody [EPR20764] (**ab214668**) at 1/1000 dilution

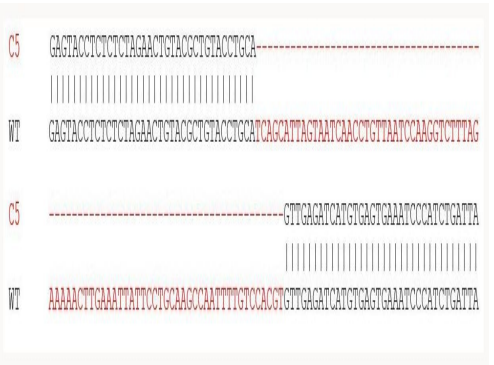
Lane 1 : Wild-type THP-1 vehicle control IFNg (0 ng/ml, 32 h), TNF-alpha (0 ng/ml, 32 h), Brefeldin A (5 ug/ml, 6 h) cell lysate
Lane 2 : Wild-type THP-1 treated IFNg (100 ng/ml, 32 h), TNF-alpha (10 ng/ml, 32 h), Brefeldin A (5 ug/ml, 6 h) cell lysate
Lane 3 : CXCL10 knockout THP-1 vehicle control IFNg (0 ng/ml, 32 h), TNF-alpha (0 ng/ml, 32 h), Brefeldin A (5 ug/ml, 6 h) cell lysate
Lane 4 : CXCL10 knockout THP-1 treated IFN-gamma (100 ng/ml, 32 h), TNF-alpha (10 ng/ml, 32 h), Brefeldin A (5 ug/ml, 6 h) cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 10 kDa
Observed band size: 11 kDa

False colour image of Western blot: Anti-IP10 antibody [EPR20764] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (**ab7291**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab214668** was shown to bind specifically to IP10. A band was observed at 11 kDa in treated wild-type THP-1 cell lysates with no signal observed at this size in treated CXCL10 knockout cell line ab277860 (knockout cell lysate **ab282997**). To generate this image, wild-type and CXCL10 knockout THP-1 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) at 1/20000 dilution.



Sanger Sequencing - Human CXCL10 (IP10) knockout THP-1 cell line (ab277860)

80 bp Deletion in Exon 2

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