abcam

Product datasheet

Human TMED10 (TMP21) knockout HEK-293T cell line ab266227

4 Images

Overview

Product name	Human TMED10 (TMP21) knockout HEK-293T cell line
Parental Cell Line	HEK293T
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 2 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	**Recommended control:** Human wild-type HEK293T cell line (**ab255449**). 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:** DMEM (High Glucose) + 10% FBS **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 $2x10^4$ cells/cm^2. 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. **Subculture guidelines:** All seeding densities should be based on cell counts gained by established methods. A guide seeding density of $2x10^4$ cells/cm^2 is recommended.

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, ERS Genomics Limited and Sigma-Aldrich Co. LLC, and is developed with patented technology. For full details of the licenses and 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	Adherent
Tissue	Kidney
Cell type	epithelial
STR Analysis	Amelogenin X D5S818: 8, 9 D13S317: 12, 14 D7S820: 11 D16S539: 9, 13 vWA: 16, 19 TH01: 7, 9.3 TPOX: 11 CSF1PO: 11, 12
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	Involved in vesicular protein trafficking. Mainly functions in the early secretory pathway. Thought to act as cargo receptor at the lumenal side for incorporation of secretory cargo molecules into transport vesicles and to be involved in vesicle coat formation at the cytoplasmic side. In COPII vesicle-mediated anterograde transport involved in the transport of GPI-anchored proteins and proposed to act togther with TMED2 as their cargo receptor; the function specifically implies SEC24C and SEC24D of the COPII vesicle coat and lipid raft-like microdomains of the ER. Recognizes GPI anchors structural remodeled in the ER by PGAP1 and MPPE1 (By similarity). In COPI vesicle-mediated retrograde transport involved in the biogenesis of COPI vesicles and vesicle coat recruitment. On Golgi membranes, acts as primary receptor for ARF1-GDP which is involved in COPI-vesicle formation. Increases coatomer-dependent GTPase-activating activity of ARFGAP2. Involved in trafficking of G protein-coupled receptors (GPCRs). Regulates F2LR1, OPRM1 and P2RY4 exocytic trafficking from the Golgi to the plasma membrane thus contributing to receptor resensitization. Involved in trafficking of amyloid beta A4 protein and soluble APP-beta release (independent of modulation of gamma-secretase activity). As part of the presenilin-dependent gamma-secretase complex regulates gamma-cleavages of the amyloid beta A4 protein to yield amyloid-beta 40 (Abeta40). Involved in organization of the Golgi apparatus.
Tissue specificity	Ubiquitous.
Sequence similarities	Belongs to the EMP24/GP25L family. Contains 1 GOLD domain.
Domain	The lumenal domain mediates localization to the plasma membrane by partially overriding the ER retention by the cytoplasmic domain.
Cellular localization	Golgi apparatus > cis-Golgi network membrane. Melanosome. Endoplasmic reticulum membrane. Endoplasmic reticulum-Golgi intermediate compartment membrane. Cytoplasmic

vesicle > secretory vesicle membrane. Cell membrane. Golgi apparatus > trans-Golgi network membrane. Identified by mass spectrometry in melanosome fractions from stage I to stage IV. Cycles between compartments of the early secretatory pathway.

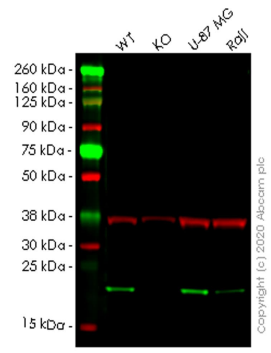
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab266227 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: 25 kDa.

Images



Western blot - Human TMED10 knockout HEK293T cell line (ab266227)

All lanes : Anti-TMP21 antibody [EPR9036(B)] (**ab134948**) at 1/1000 dilution

Lane 1 : Wild-type HEK293T cell lysate
Lane 2 : TMED10 knockout HEK293T cell lysate
Lane 3 : U-87 MG cell lysate
Lane 4 : Raji cell lysate

Lysates/proteins at 20 µg per lane.

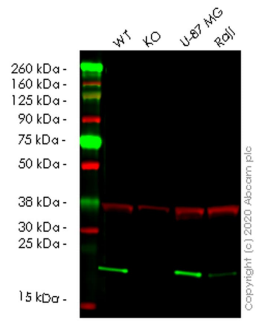
Secondary
All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/10000 dilution

Predicted band size: 25 kDa
Observed band size: 19 kDa

Lanes 1-4: Merged signal (red and green). Green - **ab134948** observed at 19 kDa. Red - loading control **ab8245** observed at 36 kDa.

ab134948 Anti-TMP21 antibody [EPR9036(B)] was shown to specifically react with TMP21 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266227 (knockout cell lysate **ab258233**) was used. Wild-type and TMP21 knockout samples were subjected to SDS-PAGE. **ab134948** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated

at room temperature for 2.5 hours 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.



Western blot - Human TMED10 knockout HEK293T cell line (ab266227)

All lanes : Anti-TMP21 antibody [EPR9037(B)] (**ab133771**) at 1/1000 dilution

Lane 1 : Wild-type HEK293T cell lysate
Lane 2 : TMED10 knockout HEK293T cell lysate
Lane 3 : U-87 MG cell lysate
Lane 4 : Raji cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/10000 dilution

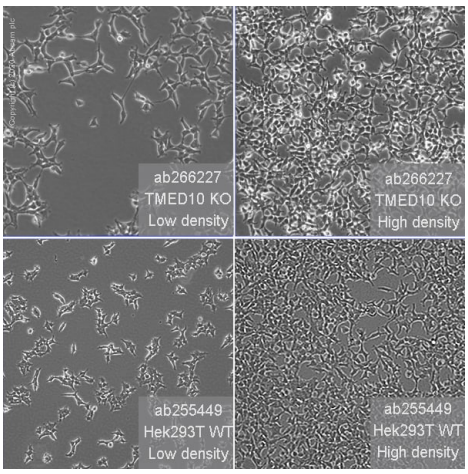
Predicted band size: 25 kDa
Observed band size: 19 kDa

Lanes 1-4: Merged signal (red and green). Green - **ab133771** observed at 19 kDa. Red - loading control **ab8245** observed at 36 kDa.

ab133771 Anti-TMP21 antibody [EPR9037(B)] was shown to specifically react with TMP21 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266227 (knockout cell lysate **ab258233**) was used. Wild-type and TMP21 knockout samples were subjected to SDS-PAGE. **ab133771** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated at room temperature for 2.5 hours 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.

Sanger Sequencing - Human TMED10 knockout HEK293T cell line (ab266227)

Homozygous: 2 bp insertion in exon1



Human TMED10 (TMP21) knockout HEK293T cell line (ab266227)

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

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.com/abpromise** or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors