abcam

Product datasheet

Human MYOF (Myoferlin) knockout HeLa cell line ab265782

画像数 2

製品の概要

製品名	Human MYOF (Myoferlin) knockout HeLa cell line		
Parental Cell Line	HeLa		
Organism	Human		
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 10 bp deletion in exon 6		
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	Recommended control: Human wild-type HeLa cell line (<u>ab255928</u>). 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.		
	 Thaw the vial in 37°C water bath for approximately 1-2 minutes. 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. 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 2x10⁴ cells/cm². Seeding density is given as a guide only and should be scaled to align with individual lab schedules. Incubate the culture at 37°C incubator with 5% CO₂. 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 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.

製品の特性

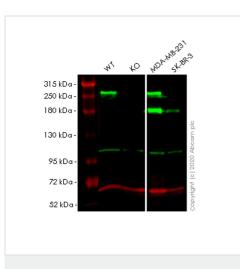
Number of cells	1 x 10 ⁶ cells/vial, 1 mL		
Adherent /Suspension	Adherent		
Tissue	Cervix		
Cell type	epithelial		
Disease	Adenocarcinoma		
Gender	Female		
STR Analysis	Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 vWA: 16, 18 TH01: 7 TPOX: 8, 12 CSF1PO: 9, 10		
Antibiotic resistance	Puromycin 1.00µg/ml		
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			
機能	Calcium/phospholipid-binding protein that plays a role in the plasmalemma repair mechanism of endothelial cells that permits rapid resealing of membranes disrupted by mechanical stress. Involved in endocytic recycling. Implicated in VEGF signal transduction by regulating the levels of the receptor KDR.		
組織特異性	Expressed in myoblast and endothelial cells (at protein level). Highly expressed in cardiac and skeletal muscles. Also present in lung, and at very low levels in kidney, placenta and brain.		
配列類似性	Belongs to the ferlin family. Contains 5 C2 domains.		
ドメイン	The C2 domain 1 associates with lipid membranes in a calcium-dependent manner.		
細胞内局在	Cell membrane. Nucleus membrane. Cytoplasmic vesicle membrane. Concentrated at the membrane sites of both myoblast-myoblast and myoblast-myotube fusions. Detected at the plasmalemma in endothelial cells lining intact blood vessels (By similarity). Found at nuclear and plasma membranes. Enriched in undifferentiated myoblasts near the plasma membrane in puncate structures.		
	puncate structures.		

アプリケーション

The Abpromise guaranteeAbpromise保証は、次のテスト済みアプリケーションにおけるab265782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: 235 kDa.

画像



Western blot - Human MYOF knockout HeLa cell line (ab265782) All lanes : Anti-Myoferlin antibody [EPR18887] (<u>ab178386</u>) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate Lane 2 : MYOF knockout HeLa cell lysate Lane 3 : MDA-MB-231 cell lysate Lane 4 : SK-BR-3 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) at 1/10000 dilution

Predicted band size: 235 kDa Observed band size: 180,250 kDa

Lanes 1-4: Merged signal (red and green). Green - <u>ab178386</u> observed at 250,180 kDa. Red - loading control <u>ab7291</u> observed at 50 kDa.

<u>ab178386</u> Anti-Myoferlin antibody [EPR18887] was shown to specifically react with Myoferlin in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab265782 (knockout cell lysate <u>ab257547</u>) was used. Wild-type and Myoferlin knockout samples were subjected to SDS-PAGE. <u>ab178386</u> and Anti-alpha Tubulin antibody [DM1A] - Loading Control (<u>ab7291</u>) 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 (<u>ab216773</u>) and Goat anti-Mouse IgG H&L (IRDye[®] 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging. Homozygous: 10 bp deletion in exon 6.

Sanger Sequencing - Human MYOF knockout HeLa cell line (ab265782)

GCTCGGAGGCTCACCAAAGT-----CGGCGGATGCTGTCAAATAAGCCACAGGAC

GCTCGGAGGCTCACCAAAGTAAAGAACAGCCGGCGGATGCTGTCAAATAAGCCACAGGAC

Mut

WT

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