abcam

Product datasheet

Human ULK1 knockout A549 cell line ab277825

Overview

Product name	Human ULK1 knockout A549 cell line
Parental Cell Line	A549
Organism	Human
Passage number	<20
Biosafety level	1
General notes	**Recommended control:** Human wild-type A549 cell line (ab275463). 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:** F-12K + 10% FBS **Initial handling guidelines:** Upon arrival, the vial should be stored in liquid nitrogen vapor phase and not at -80ºC. Storage at -80ºC may result in loss of viability. 1. Thaw the vial in 37ºC water bath 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 (Click here to view haemocytometer protocol) 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. This should allow for confluency within 48 hours. Seeding density is given as a guide only and should be scaled to align with individual lab schedules. 4. Incubate the culture at 37ºC 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 for confluency (80-90% confluence) within 48 hours. 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. Click here to view the Mammalian cell tissue culture protocol

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Properties

Number of cells	1 x 10^6 cells/vial, 1 mL
Viability	~90%
Adherent /Suspension	Adherent
Tissue	Lung
Cell type	epithelial
Disease	Carcinoma
Gender	Male
Mycoplasma free	Yes
Storage instructions	Shipped on Dry Ice. Store in liquid nitrogen.
Storage buffer	Constituents: 8.7% DMSO, 2% Cellulose, methyl ether

Target

Function	Serine/threonine-protein kinase involved in autophagy in response to starvation. Acts upstream of phosphatidylinositol 3-kinase PIK3C3 to regulate the formation of autophagophores, the precursors of autophagosomes. Part of regulatory feedback loops in autophagy: acts both as a downstream effector and negative regulator of mammalian target of rapamycin complex 1 (mTORC1) via interaction with RPTOR. Activated via phosphorylation by AMPK and also acts as a regulator of AMPK by mediating phosphorylation of AMPK subunits PRKAA1, PRKAB2 and PRKAG1, leading to negatively regulate AMPK activity. May phosphorylate ATG13/KIAA0652 and RPTOR; however such data need additional evidences. Plays a role early in neuronal differentiation and is required for granule cell axon formation. May also phosphorylate SESN2 and SQSTM1 to regulate autophagy (PubMed:25040165).
Tissue specificity	Ubiquitously expressed. Detected in the following adult tissues: skeletal muscle, heart, pancreas, brain, placenta, liver, kidney, and lung.
Sequence similarities	Belongs to the protein kinase superfamily. Ser/Thr protein kinase family. APG1/unc-51/ULK1 subfamily. Contains 1 protein kinase domain.
Post-translational modifications	Autophosphorylated. Phosphorylated under nutrient-rich conditions; dephosphorylated during starvation or following treatment with rapamycin. Under nutrient sufficiency, phosphorylated by MTOR/mTOR, disrupting the interaction with AMPK and preventing activation of ULK1 (By similarity). In response to nutrient limitation, phosphorylated and activated by AMPK, leading to activate autophagy.
Cellular localization	Cytoplasm, cytosol. Preautophagosomal structure. Under starvation conditions, is localized to puncate structures primarily representing the isolation membrane that sequesters a portion of the cytoplasm resulting in the formation of an autophagosome.

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