abcam

Product datasheet

# Human MET knockout A549 cell line ab286606

2 Images

## Overview

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| **Product name** | Human MET knockout A549 cell line |
| **Parental Cell Line** | A549 |
| **Organism** | Human |
| **Passage number** | <20 |
| **Knockout validation** | Western Blot (WB) |
| **Biosafety level** | 1 |
| **General notes** | Although we aim to provide customers with a homozygous clone, feasibility will be dependent on the biology of the protein. Should only heterozygous edits be achieved, you will be notified of the outcome and be asked to confirm whether the cell line is acceptable. All clones will be accompanied with DNA sequencing data, and the mutation description.<br><br>**Recommended control**: Human wild-type A549 cell line (**ab288558**). 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.<br><br>**Cryopreservation cell medium**: Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose.<br><br>**Culture medium**: F-12K + 10% FBS<br><br>**Initial handling guidelines**:<br>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.<br>1. Thaw the vial in 37°C water bath for approximately 1-2 minutes.<br>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.<br>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 2x103-1x10$^4$ cells/cm2. Seeding density is given as a guide only and should be scaled to align with individual lab schedules.<br>4. 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 $6x10^4$ cells/cm2 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.
Do not allow the cell density to exceed $7x10^4$ cells/cm2.
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

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| **Number of cells** | 1000000 cells/vial, 1 mL |
| **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% Dimethylsulfoxide, 2% Cellulose, methyl ether |

## Target

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| **Function** | Receptor for hepatocyte growth factor and scatter factor. Has a tyrosine-protein kinase activity. Functions in cell proliferation, scattering, morphogenesis and survival. |
| **Involvement in disease** | Note=Activation of MET after rearrangement with the TPR gene produces an oncogenic protein.<br>Note=Defects in MET may be associated with gastric cancer.<br>Defects in MET are a cause of hepatocellular carcinoma (HCC) [MIM:114550].<br>Defects in MET are a cause of renal cell carcinoma papillary (RCCP) [MIM:605074]. It is a subtype of renal cell carcinoma tending to show a tubulo-papillary architecture formed by numerous, irregular, finger-like projections of connective tissue. Renal cell carcinoma is a heterogeneous group of sporadic or hereditary carcinoma derived from cells of the proximal renal tubular epithelium. It is subclassified into common renal cell carcinoma (clear cell, non-papillary carcinoma), papillary renal cell carcinoma, chromophobe renal cell carcinoma, collecting duct carcinoma with medullary carcinoma of the kidney, and unclassified renal cell carcinoma.<br>Note=A common allele in the promoter region of the MET shows genetic association with susceptibility to autism in some families. Functional assays indicate a decrease in MET promoter activity and altered binding of specific transcription factor complexes.<br>Note=MET activating mutations may be involved in the development of a highly malignant, metastatic syndrome known as cancer of unknown primary origin (CUP) or primary occult malignancy. Systemic neoplastic spread is generally a late event in cancer progression. However, in some instances, distant dissemination arises at a very early stage, so that metastases reach |

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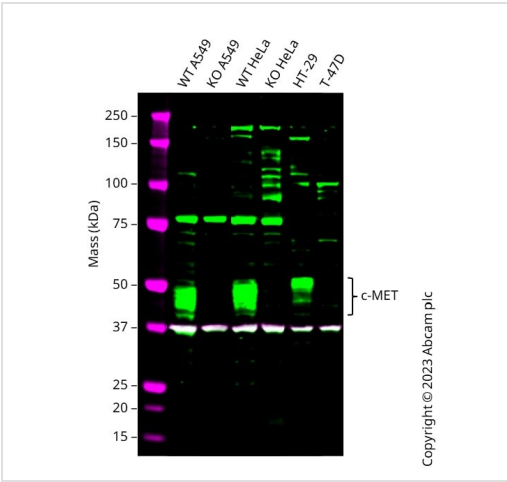
| | clinical relevance before primary lesions. Sometimes, the primary lesions cannot be identified in spite of the progresses in the diagnosis of malignancies. |
| **Sequence similarities** | Belongs to the protein kinase superfamily. Tyr protein kinase family.<br>Contains 3 IPT/TIG domains.<br>Contains 1 protein kinase domain.<br>Contains 1 Sema domain. |
| **Domain** | The kinase domain is involved in SPSB1 binding. |
| **Post-translational modifications** | Dephosphorylated by PTPRJ at Tyr-1349 and Tyr-1365. |
| **Cellular localization** | Membrane. |

## Images




Western blot - Human MET knockout A549 cell line (ab286606)

**All lanes :** Anti-Met (c-Met) antibody [EP1454Y] - N-terminal (**ab51067**) at 1/1000 dilution

**Lane 1 :** Wild-type A549 cell lysate
**Lane 2 :** MET knockout A549 cell lysate
**Lane 3 :** Wild-type HeLa **ab255929** cell lysate
**Lane 4 :** MET (Met (c-Met)) knockout HeLa **ab256991** cell lysate
**Lane 5 :** HT-29 cell lysate
**Lane 6 :** T-47D cell lysate

Lysates/proteins at 20 µg per lane.

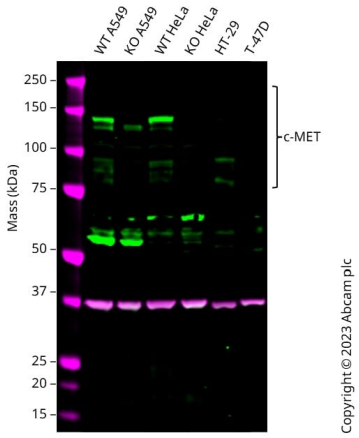
Developed using the ECL technique.

Performed under reducing conditions.

**Observed band size:** 40-50 kDa

Western blot: Anti-MET antibody [EP1454Y] (**ab51067**) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (**ab8245**) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, **ab51067** was shown to bind specifically to MET. A band was observed at 40-50 kDa in wild-type A549 cell lysates with no signal observed at this size in MET knockout cell line. To generate this image, wild-type and MET knockout A549 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 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.




Western blot - Human MET knockout A549 cell line (ab286606)

**All lanes :** Anti-Met (c-Met) antibody [EPR19554-110] (**ab254252**) at 1/1000 dilution

**Lane 1 :** Wild-type A549 cell lysate
**Lane 2 :** MET knockout A549 cell lysate
**Lane 3 :** Wild-type HeLa **ab255929** cell lysate
**Lane 4 :** MET (Met (c-Met)) knockout HeLa **ab256991** cell lysate
**Lane 5 :** HT-29 cell lysate
**Lane 6 :** T-47D cell lysate

Lysates/proteins at 20 µg per lane.

Developed using the ECL technique.

Performed under reducing conditions.

**Observed band size:** 80-140 kDa

Western blot: Anti-MET antibody [EPR19554-110] (**ab254252**) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (**ab8245**) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, **ab254252** was shown to bind specifically to MET. A band was observed at 80-140 kDa in wild-type A549 cell lysates with no signal observed at this size in MET knockout cell line. To generate this image, wild-type and MET knockout A549 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 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.

The full length MET protein is glycosylated and has multiple cleavage sites, so cleaved products can be detected between ~40-140 kDa

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

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