abcam

Product datasheet

# Human RRM2B (p53R2) knockout HeLa cell line ab261769

画像数 3

## 製品の概要

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| **製品名** | Human RRM2B (p53R2) knockout HeLa cell line |
| **Parental Cell Line** | HeLa |
| **Organism** | Human |
| **Mutation description** | Knockout achieved by using CRISPR/Cas9, 1 bp deletion in exon 1 and Insertion of the selection cassette in exon 1 |
| **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 (**ab255448**). 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:** DMEM (High Glucose) + 10% FBS<br><br>**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.<br><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 $2x10^4$ cells/$cm^2$. 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% $CO_2$. Cultures should be monitored daily.<br><br>**Subculture guidelines:**<br><br>All seeding densities should be based on cell counts gained by established methods.<br>A guide seeding density of $2x10^4$ cells/$cm^2$ is recommended.<br>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.
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We will provide viable cells that proliferate on revival.

## 製品の特性

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| **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 |
| **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

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| **機能** | Plays a pivotal role in cell survival by repairing damaged DNA in a p53/TP53-dependent manner. Supplies deoxyribonucleotides for DNA repair in cells arrested at G1 or G2. Contains an iron-tyrosyl free radical center required for catalysis. Forms an active ribonucleotide reductase (RNR) complex with RRM1 which is expressed both in resting and proliferating cells in response to DNA damage. |
| **組織特異性** | Widely expressed at a high level in skeletal muscle and at a weak level in thymus. Expressed in epithelial dysplasias and squamous cell carcinoma. |
| **パスウェイ** | Genetic information processing; DNA replication. |
| **関連疾患** | Defects in RRM2B are the cause of mitochondrial DNA depletion syndrome type 8A (MTDPS8A) [MIM:612075]. A disorder due to mitochondrial dysfunction characterized by various combinations of neonatal hypotonia, neurological deterioration, respiratory distress, lactic acidosis, and renal tubulopathy.<br>Defects in RRM2B are the cause of mitochondrial DNA depletion syndrome type 8B (MTDPS8B) [MIM:612075]. A disease due to mitochondrial dysfunction and characterized by ophthalmoplegia, ptosis, gastrointestinal dysmotility, cachexia, peripheral neuropathy.<br>Defects in RRM2B are the cause of progressive external ophthalmoplegia with mitochondrial DNA deletions autosomal dominant type 5 (PEOA5) [MIM:613077]. A disorder characterized by progressive weakness of ocular muscles and levator muscle of the upper eyelid. In a minority of cases, it is associated with skeletal myopathy, which predominantly involves axial or proximal muscles and which causes abnormal fatigability and even permanent muscle weakness. Ragged-red fibers and atrophy are found on muscle biopsy. A large proportion of chronic ophthalmoplegias are associated with other symptoms, leading to a multisystemic pattern of this |

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| | disease. Additional symptoms are variable, and may include cataracts, hearing loss, sensory axonal neuropathy, ataxia, depression, hypogonadism, and parkinsonism. |
| **配列類似性** | Belongs to the ribonucleoside diphosphate reductase small chain family. |
| **細胞内局在** | Cytoplasm. Nucleus. Translocates from cytoplasm to nucleus in response to DNA damage. |

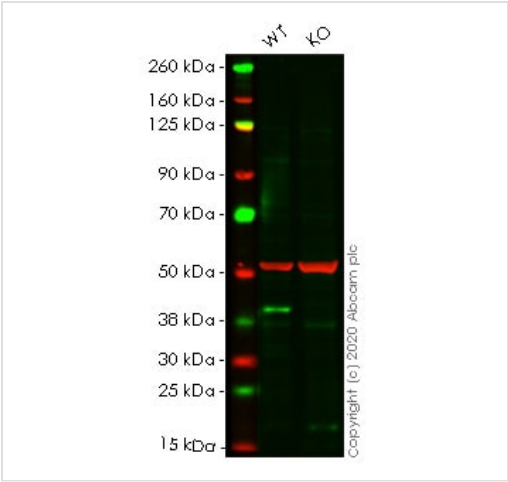
## アプリケーション

**The Abpromise guarantee** **Abpromise保証は、次のテスト済みアプリケーションにおけるab261769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: 40 kDa. |

## 画像




Western blot - Human RRM2B (p53R2) knockout HeLa cell line (ab261769)

**All lanes :** Anti-p53R2 antibody [EPR8816] (**ab154194**) at 1/1000 dilution

**Lane 1 :** Wild-type HeLa cell lysate
**Lane 2 :** RRM2B knockout HeLa cell lysate

Lysates/proteins at 40 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 40 kDa
**Observed band size:** 40 kDa

**Lanes 1- 2:** Merged signal (red and green). Green - **ab154194** observed at 40 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) observed at 50 kDa.

**ab154194** was shown to react with p53R2 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab261769 (knockout cell lysate **ab257215**) was used. Wild-type HeLa and RRM2B knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab154194** and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) overnight at 4°C at a 1 in 1000 dilution and a 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 RRM2B knockout HeLa cell line (ab261769)

Allele-1: 1 bp deletion in exon 1.


Sanger Sequencing - Human RRM2B knockout HeLa cell line (ab261769)

Allele-2: Insertion of the selection cassette in exon 1.

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