

# Human PKN1 knockout HEK-293T cell line ab266599

画像数 4

## 製品の概要

製品名	Human PKN1 knockout HEK-293T cell line
Parental Cell Line	HEK293T
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 1 bp insertion in exon 2 and Insertion of the selection cassette in exon 2
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	<p><b>Recommended control:</b> Human wild-type HEK293T cell line (<a href="#">ab255449</a>). 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.</p> <p><b>Cryopreservation cell medium:</b> Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose.</p> <p><b>Culture medium:</b> DMEM (High Glucose) + 10% FBS</p> <p><b>Initial handling guidelines:</b> 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.</p> <ol style="list-style-type: none"> <li>1. Thaw the vial in 37°C water bath for approximately 1-2 minutes.</li> <li>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.</li> <li>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 <math>2 \times 10^4</math> cells/cm<sup>2</sup>. Seeding density is given as a guide only and should be scaled to align with individual lab schedules.</li> <li>4. Incubate the culture at 37°C incubator with 5% CO<sub>2</sub>. Cultures should be monitored daily.</li> </ol> <p><b>Subculture guidelines:</b></p> <p>All seeding densities should be based on cell counts gained by established methods. A guide seeding density of <math>2 \times 10^4</math> cells/cm<sup>2</sup> is recommended.</p> <p>A partial media change 24 hours prior to subculture may be helpful to encourage growth, if</p>

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

Number of cells	1 x 10 <sup>6</sup> 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
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	PKC-related serine/threonine-protein kinase involved in various processes such as regulation of the intermediate filaments of the actin cytoskeleton, cell migration, tumor cell invasion and transcription regulation. Regulates the cytoskeletal network by phosphorylating proteins such as VIM and neurofilament proteins NEFH, NEFL and NEFM, leading to inhibit their polymerization. Phosphorylates 'Ser-575', 'Ser-637' and 'Ser-669' of MAPT/Tau, lowering its ability to bind to microtubules, resulting in disruption of tubulin assembly. Acts as a key coactivator of androgen receptor (ANDR)-dependent transcription, by being recruited to ANDR target genes and specifically mediating phosphorylation of 'Thr-11' of histone H3 (H3T11ph), a specific tag for epigenetic transcriptional activation that promotes demethylation of histone H3 'Lys-9' (H3K9me) by KDM4C/JMJD2C. Phosphorylates HDAC5, HDAC7 and HDAC9, leading to impair their import in the nucleus. Phosphorylates 'Thr-38' of PPP1R14A, 'Ser-159', 'Ser-163' and 'Ser-170' of MARCKS, and GFAP. Able to phosphorylate RPS6 in vitro.
組織特異性	Found ubiquitously. Expressed in heart, brain, placenta, lung, skeletal muscle, kidney and pancreas. Expressed in numerous tumor cell lines, especially in breast tumor cells.
配列類似性	Belongs to the protein kinase superfamily. AGC Ser/Thr protein kinase family. PKC subfamily. Contains 1 AGC-kinase C-terminal domain. Contains 1 C2 domain. Contains 1 protein kinase domain. Contains 3 REM (Hr1) repeats.
ドメイン	The C1 domain does not bind the diacylglycerol (DAG).
翻訳後修飾	Autophosphorylated; preferably on serine. Phosphorylated during mitosis. Activated by limited proteolysis with trypsin.
細胞内局在	Cytoplasm. Nucleus. Endosome. Cell membrane. Cleavage furrow. Midbody. Associates with chromatin in a ligand-dependent manner. Localization to endosomes is mediated via its

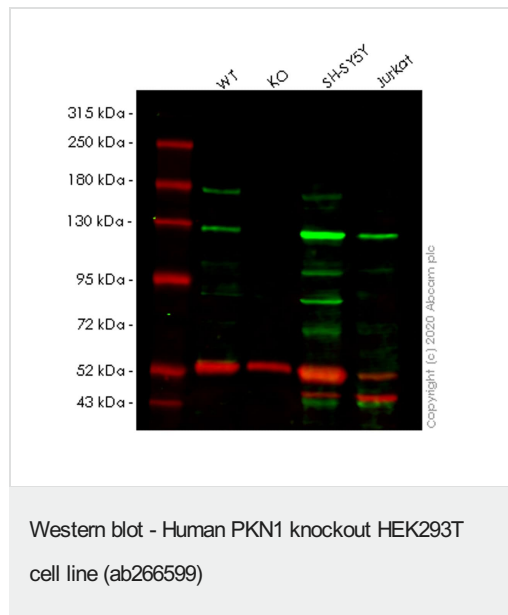
interaction with RHOB. Association to the cell membrane is dependent on Ser-374 phosphorylation. Accumulates during telophase at the cleavage furrow and finally concentrates around the midbody in cytokinesis.

## 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ab266599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: 104 kDa.

## 画像



**All lanes :** Anti-PKN antibody [EPR3237] ([ab108976](#)) at 1/1000 dilution

**Lane 1 :** Wild-type HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate

**Lane 2 :** PKN1 knockout HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate

**Lane 3 :** SH-SY5Y (Human neuroblastoma cell line from bone marrow) whole cell lysate

**Lane 4 :** Jurkat (Human T cell leukemia cell line from peripheral blood) whole 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:** 104 kDa

**Observed band size:** 120 kDa

**Lanes 1-4:** Merged signal (red and green). Green - [ab108976](#) observed at 120 kDa. Red - loading control [ab7291](#) observed at 50 kDa.

[ab108976](#) Anti-PKN antibody [EPR3237] was shown to specifically react with PKN in wild-type HEK-293T cells. Loss of

signal was observed when knockout cell line ab266599 (knockout cell lysate **ab258586**) was used. Wild-type and PKN knockout samples were subjected to SDS-PAGE. **ab108976** and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) 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 (**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.

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Mut  TGGAGCGGAGCGGCTGCGGCGGGAATCCGCAAGGAGCTGAAGCTGAAGGGGTGCT
      ||||||||||||||||||||||||||||||||||||||||||||||||||||||
WT   TGGAGCGGAGCGGCTGCGGCGGGAATCC GCAAGGAGCTGAAGCTGAAGGGGTGCT
  
```

Sanger Sequencing - Human PKN1 knockout  
HEK293T cell line (ab266599)

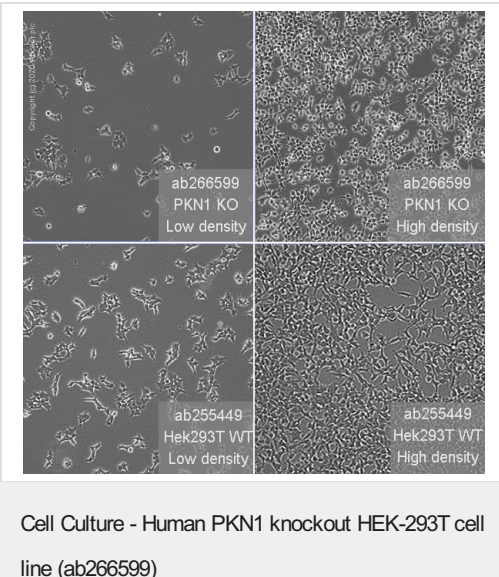
Allele-1: 1 bp insertion in exon2

```

Mut  GCGGCTGCGGCGGGAATCC*****Insertion*****GCAAGGAGCTGAAGCTGAAG
      ||||||||||||||||||                               ||||||||||||||||||
WT   GCGGCTGCGGCGGGAATCC                               GCAAGGAGCTGAAGCTGAAG
  
```

Sanger Sequencing - Human PKN1 knockout  
HEK293T cell line (ab266599)

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



Representative images PKN1 knockout HEK293T cells, low and high confluency examples (top left and right respectively) and wild-type HEK293T cells, low and high confluency (bottom left and right respectively) showing typical adherent, epithelial-like morphology. Images were captured at 10X magnification using a EVOS M5000 microscope.

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