abcam

Product datasheet

Human TPBG knockout MCF7 cell lysate ab269661

画像数 4

製品の概要

製品名 Human TPBG knockout MCF7 cell lysate

製品の概要 Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line MCF7
Organism Human

Mutation description Knockout achieved by CRISPR/Cas9; X = 1 bp insertion; Frameshift: 97%

Passage number <20

Knockout validation Next Generation Sequencing (NGS), Western Blot (WB)

 $\label{eq:Reconstitution notes} \textbf{To use as WB control, resuspend the lyophilizate in 50 μL of LDS* Sample Buffer to have a final μL of LDS* Sample$

concentration of 2 mg/ml. For reducing conditions, we recommend a final concentration of 0.1 M

DTT.

*Usage of SDS sample buffer is not recommended with these lyophilized lysates.

特記事項

Lysate preparation: Our lysates are made using RIPA buffer to which we add a protease inhibitor cocktail and phosphatase inhibitor cocktail (ratio: 300:100:10). *This means that the protein of interest is denatured.* If you require a native form of the protein please use the live cell version - found **here**. Please refer to our lysis protocol for further details on how our lysates are prepared.

User storage instructions: Lyophilizate may be stored at 4°C. After reconstitution, store at -20°C for short-term storage or -80°C for long-term storage.

Access thousands of knockout cell lysates, generated from commonly used cancer cell lines. **See here for more information on knockout cell lysates.**

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アプリケーション **適用あり**: WB

保存方法

Store at -80°C. Please refer to protocols.

	1 kit
ab280536 - Human TPBG knockout MCF7 cell lysate	1 x 100μg
ab269599 - Human wild-type MCF7 cell lysate	1 x 100μg

Cell type epithelial

Disease Adenocarcinoma

Gender Female

ターゲット情報

組織特異性 Expressed by all types of trophoblasts as early as 9 weeks of development. Specific for

trophoblastic cells except for amniotic epithelium. In adult tissues, the expression is limited to a

few epithelial cell types but is found on a variety of carcinoma.

配列類似性 Contains 6 LRR (leucine-rich) repeats.

Contains 1 LRRCT domain. Contains 1 LRRNT domain.

細胞内局在 Membrane.

製品の状態 5T4 is a 72 kDa transmembrane glycoprotein that exhibits restricted expression in human and

mouse adult tissues, is upregulated on many carcinomas and tumour expression correlates with poorer clinical outcome in ovarian, gastric and colorectal cancers. mES cells lack cell surface expression of the 5T4 antigen, and both protein and mRNA are rapidly upregulated following induction of differentiation. This proteins expression correlates with downregulation of OCT-4 and

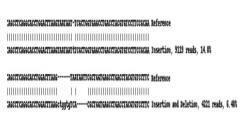
Tra-1-60 (Ward et al., 2006).

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab269661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。

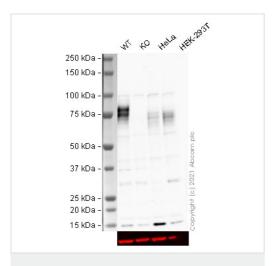
アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration. Predicted molecular weight: 46 kDa.

画像



Next Generation Sequencing - Human TPBG knockout MCF7 cell lysate (ab269661)

Frameshift: 97%



Western blot - Human PCBG knockout MCF7 cell lysate

Lane 1: Wild-type MCF7 (Human breast adenocarcinoma cell line) whole cell lysate, 20 ug

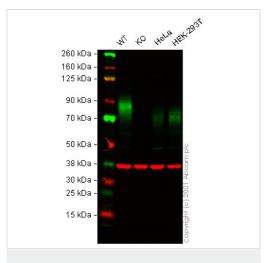
Knockout achieved by CRISPR/Cas9; X = 1 bp insertion;

Lane 2: TPBG knockout MCF7 (Human breast adenocarcinoma cell line) whole cell lysate, 20 ug

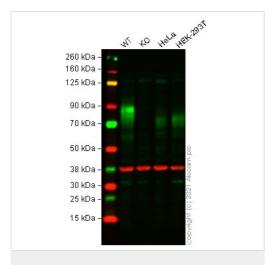
Lane 3: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate, 20 ug

Lane 4: HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate, 20 ug

ab199547 was shown to react with 5T4 (HRP) in wild-type MCF7 cells in western blot. Loss of signal was observed when TPBG knockout cell line ab269499 (knockout cell lysate ab269661) was used. Membranes were blocked in 3 % milk in TBS-T (0.1 %Tween®) before incubation with ab199547 overnight at 4 °C at a 1 in 5000 dilution Blots were developed with Optiblot ECL reagent (ab133456) and imaged.



Western blot - Human TPBG knockout MCF7 cell lysate



Western blot - Human TPBG knockout MCF7 cell lysate

Lane 1: Wild-type MCF7 (Human breast adenocarcinoma cell line) whole cell lysate, 20 ug

Lane 2: TPBG knockout MCF7 (Human breast adenocarcinoma cell line) whole cell lysate, 20 ug

Lane 3: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate, 20 ug

Lane 4: HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate, 20 ug

Lanes 1 - 4: Merged signal (red and green). Green - <u>ab134162</u> observed at 85 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab134162 was shown to react with 5T4 in wild-type MCF7 cells in Western blot with loss of signal observed in TPBG knockout cell line ab269499 (knockout cell lysate ab269661). Wild-type MCF7 and TPBG knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in before incubation with ab134162 and ab8245 (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.

Lane 1: Wild-type MCF7 (Human breast adenocarcinoma cell line) whole cell lysate, 20 ug

Lane 2: TPBG knockout MCF7 (Human breast adenocarcinoma cell line) whole cell lysate, 20 ug

Lane 3: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate, 20 ug

Lane 4: HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate, 20 ug

Lanes 1 - 4: Merged signal (red and green). Green - <u>ab129058</u> observed at 85 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab129058 was shown to react with 5T4 in wild-type MCF7 cells in Western blot with loss of signal observed in TPBG knockout cell line ab269499 (knockout cell lysate ab269661). Wild-type MCF7 and TPBG knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in before incubation with ab129058 and ab8245 (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse lgG H&L

(IRDye[®] 680RD) preabsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.

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