abcam

Product datasheet

Human CBFB knockout A-431 cell lysate ab270495

4 图像

概述

产品概述 Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line A431
Organism Human

Mutation description Knockout achieved by CRISPR/Cas9; X = 19 bp deletion; Frameshift = 99.99%

Passage number <20

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

Reconstitution notesTo use as WB control, resuspend the lyophilizate in 50 μL of LDS* Sample Buffer to have a final

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.

说明

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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经测试应用 适用于: WB

存放说明

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

组 件	1 kit
ab280548 - Human CBFB knockout A-431 cell lysate	1 x 100μg
ab263973 - Human wild-type A-431 cell lysate	1 x 100µg

Cell type epithelial

Disease Epidermoid Carcinoma

Gender Female

靶标

功能 CBF binds to the core site, 5'-PYGPYGGT-3', of a number of enhancers and promoters, including

murine leukemia virus, polyomavirus enhancer, T-cell receptor enhancers, LCK, IL3 and GM-CSF

promoters. CBFB enhances DNA binding by RUNX1.

疾病相关 Note=A chromosomal aberration involving CBFB is associated with acute myeloid leukemia of

M4EO subtype. Pericentric inversion inv(16)(p13;q22). The inversion produces a fusion protein that consists of the 165 N-terminal residues of CBF-beta (PEPB2) with the tail region of MYH11.

序列相似性 Belongs to the CBF-beta family.

细胞定位 Nucleus.

应用

The Abpromise guarantee Ab

Abpromise™承诺保证使用ab270495于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		Use at an assay dependent concentration.

图片

STTATCTGGAAAGGCTGGATTGATCTCCAAAGACTGGATGGTATGG Reference

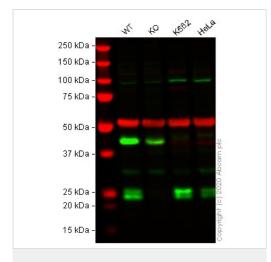
1010100001

STTATCTGGAAAGG-----CTGGATGGTATGG Deletion, 63864 reads, 95.86%

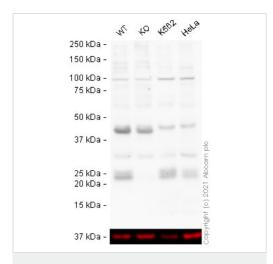
Next Generation Sequencing - Human CBFB

knockout A-431 cell lysate (ab270495)

Knockout achieved by CRISPR/Cas9; X = 19 bp deletion; Frameshift = 99.99%



Western blot - Human CBFB knockout A-431 cell lysate (ab270495)



Western blot - Human CBFB knockout A-431 cell lysate (ab270495)

Lane 1: Wild-type A431 cell lysate 20 ug

Lane 2: CBFB knockout A431 cell lysate 20 ug

Lane 3: K562 (Human chronic myelogenous leukemia lymphoblast cell line) whole cell lysate 20 ug

Lane 4: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate 20 ug

ab133600 was shown to react with CBFb in wild-type A-431 cells in western blot. Loss of signal was observed when CBFB knockout cell line ab270472 (knockout cell lysate ab270495) was used.

Membranes were blocked in 2 % BSA in TBS-T (0.1 % Tween®) before incubation with ab133600 overnight at 4°C at a 1 in 1000 dilution and ab184095 (Mouse Anti-GAPDH antibody [mAbcam 9484] - Alexa Fluor® 680) at a 1 in 1000 dilution. Blots were incubated with HRP conjugated Goat anti-Rabbit (H+L) secondary antibody at 1/5000 for 1 hour at room temperature before development with Optiblot ECL reagent (ab133456) and imaging.

Lane 1: Wild-type A431 cell lysate 20 ug

Lane 2: CBFB knockout A431 cell lysate 20 ug

Lane 3: K562 (Human chronic myelogenous leukemia lymphoblast cell line) whole cell lysate 20 ug

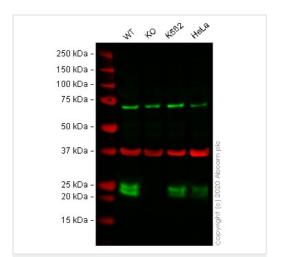
Lane 4: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate 20 ug

Lanes 1 - 4: Merged signal (red and green). Green - <u>ab133600</u> observed at 22 kDa. Red - loading control <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

<u>ab133600</u> was shown to react with CBFb in wild-type A-431 cells in western blot with loss of signal observed in CBFB knockout cell line <u>ab270472</u> (knockout cell lysate ab270495). Wild-type and CBFB knockout A-431 cell lysates were subjected to SDS-PAGE.

Membranes were blocked in 2% BSA in TBS-T (0.1% Tween[®]) before incubation with <u>ab133600</u> and <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4°C at a 1 in 1000 dilution and a 1 in

20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye[®] 800CW) preabsorbed (<u>ab216773</u>) and Goat anti-Mouse IgG H&L (IRDye[®] 680RD) preabsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Human CBFB knockout A-431 cell lysate (ab270495)

Lane 1: Wild-type A431 cell lysate 20 ug

Lane 2: CBFB knockout A431 cell lysate 20 ug

Lane 3: K562 (Human chronic myelogenous leukemia lymphoblast cell line) whole cell lysate 20 ug

Lane 4: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate 20 ug

Lanes 1 - 4: Merged signal (red and green). Green - <u>ab124693</u> observed at 24-25 kDa. Red - loading control, <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

ab124693 was shown to react with CBFb in wild-type A-431 cells in western blot Loss of signal was observed when CBFB knockout cell line ab270472 (knockout cell lysate ab270495) was used. Wild-type and CBFB knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with ab124693 and ab7291 (Mouse anti-Alpha Tubulin [DM1A] overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

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