abcam

Product datasheet

Human PDGFRB knockout SH-SY5Y cell lysate ab275523

3 Images

Overview

Product name Human PDGFRB knockout SH-SY5Y cell lysate

Product overview

Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line SHSY-5Y

Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, Homozygous: 5 bp deletion in exon 3

Passage number <20

Knockout validation Sanger Sequencing, Western Blot (WB)

Reconstitution notes To 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.

Notes

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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products that contain European Authorisation list (Annex XIV) substances.

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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.

Tested applications Suitable for: WB

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Properties

Storage instructions

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

Components	1 kit
ab277351 - Human PDGFRB knockout SHSY-5Y cell lysate	1 x 100µg
ab277350 - Human wild-type SHSY-5Y cell lysate	1 x 100µg

Cell type neuroblastoma

Disease Neuroblastoma

Gender Female

Target

Function

Receptor that binds specifically to PDGFB and PDGFD and has a tyrosine-protein kinase activity. Phosphorylates Tyr residues at the C-terminus of PTPN11 creating a binding site for the SH2 domain of GRB2.

Involvement in disease

Note=A chromosomal aberration involving PDGFRB is found in a form of chronic myelomonocytic leukemia (CMML). Translocation t(5;12)(q33;p13) with EVT6/TEL. It is characterized by abnormal clonal myeloid proliferation and by progression to acute myelogenous leukemia (AML). Note=A chromosomal aberration involving PDGFRB may be a cause of acute myelogenous leukemia. Translocation t(5;14)(q33;q32) with TRIP11. The fusion protein may be involved in clonal evolution of leukemia and eosinophilia.

Note=A chromosomal aberration involving PDGFRB may be a cause of juvenile myelomonocytic

 $leukemia.\ Translocation\ t (5;17) (q33;p11.2)\ with\ SPECC1.$

Defects in PDGFRB are a cause of myeloproliferative disorder chronic with eosinophilia (MPE) [MIM:131440]. A hematologic disorder characterized by malignant eosinophils proliferation.

Note=A chromosomal aberration involving PDGFRB is found in many instances of myeloproliferative disorder chronic with eosinophilia. Translocation t(5;12) with ETV6 on chromosome 12 creating an PDGFRB-ETV6 fusion protein.

Note=A chromosomal aberration involving PDGFRB may be the cause of a myeloproliferative disorder (MBD) associated with eosinophilia. Translocation t(1;5)(q23;q33) that forms a PDE4DIP-PDGFRB fusion protein.

Sequence similarities

Belongs to the protein kinase superfamily. Tyr protein kinase family. CSF-1/PDGF receptor subfamily.

Contains 5 lg-like C2-type (immunoglobulin-like) domains.

Contains 1 protein kinase domain.

Post-translational modifications

 $Autophosphorylated.\ Dephosphorylated\ by\ PTPRJ\ at\ Tyr-751,\ Tyr-857,\ Tyr-1009\ and\ Tyr-1021.$

Cellular localization Membrane.

Applications

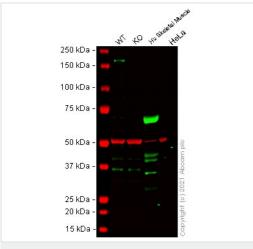
The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab275523 in the following tested applications.

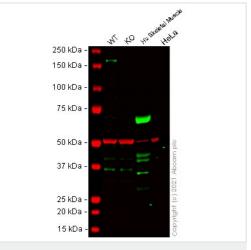
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration.

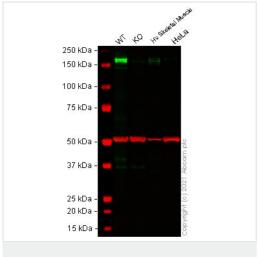
Images



cell lysate (ab275523)



Western blot - Human PDGFRB knockout SHSY-5Y



Western blot - Human PDGFRB knockout SHSY-5Y cell lysate (ab275523)

Lane 1: Wild-type SH-SY5Y cell lysate 30 ug

Lane 2: PDGFRB knockout SH-SY5Y cell lysate 30 ug

Lane 3: Human Skeletal Muscle tissue lysate 30 ug

Lane 4: HeLa cell lysate 30 ug

Lanes 1 - 4: Merged signal (red and green). Green - ab69506 observed at 170 kDa. Red - loading control ab52866 (Rabbit antialpha Tubulin antibody [EP1332Y]) observed at 55kDa.

ab69506 was shown to react with PDGFR beta in wild-type SH-SY5Y cells in Western blot with loss of signal observed in PDGFRB knockout cell line ab273749 (knockout cell lysate ab275523). Wildtype SH-SY5Y and PDGFRB knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with ab69506 and ab52866 (Rabbit anti-alpha Tubulin antibody [EP1332Y]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (ab216772) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed (ab216777) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.

Lane 1: Wild-type SH-SY5Y cell lysate 30 ug

Lane 2: PDGFRB knockout SH-SY5Y cell lysate 30 ug

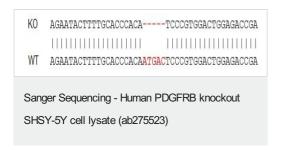
Lane 3: Human Skeletal Muscle tissue lysate 30 ug

Lane 4: HeLa cell lysate 30 ug

Lanes 1 - 4: Merged signal (red and green). Green - ab32570 observed at 170 kDa. Red - loading control ab7291 (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

ab32570 was shown to react with PDGFRB in wild-type SH-SY5Y cells in Western blot with loss of signal observed in PDGFRB knockout cell line ab273749 (knockout cell lysate ab275523). Wildtype SH-SY5Y and PDGFRB knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with ab32570 and ab7291 (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 5000 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 (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Allele-1: 5 bp deletion in exon 3

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