

Product datasheet

Human GBA knockout HeLa cell lysate ab256929

3 Images

Overview

Product name	Human GBA knockout HeLa cell lysate
Product overview	Knockout cell lysate achieved by CRISPR/Cas9.
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 1 bp insertion 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.](#)

Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of products that contain European Authorisation list (Annex XIV) substances.

It is the responsibility of our customers to check the necessity of application of REACH Authorisation, and any other relevant authorisations, for their intended uses.

This product is subject to limited use licenses from The Broad Institute, ERS Genomics Limited and Sigma-Aldrich Co. LLC, and is developed with patented technology. For full details of the licenses and patents please refer to our [limited use license](#) and [patent pages](#).

Tested applications **Suitable for:** WB

Properties

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

Components	1 kit
ab260148 - Human GBA knockout HeLa cell lysate	1 x 100µg
ab255552 - Human wild-type HeLa cell lysate	1 x 100µg

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

Target

Involvement in disease

Defects in GBA are the cause of Gaucher disease (GD) [MIM:230800]; also known as glucocerebrosidase deficiency. GD is the most prevalent lysosomal storage disease, characterized by accumulation of glucosylceramide in the reticulo-endothelial system. Different clinical forms are recognized depending on the presence (neuronopathic forms) or absence of central nervous system involvement, severity and age of onset.

Defects in GBA are the cause of Gaucher disease type 1 (GD1) [MIM:230800]; also known as adult non-neuronopathic Gaucher disease. GD1 is characterized by hepatosplenomegaly with consequent anemia and thrombopenia, and bone involvement. The central nervous system is not involved.

Defects in GBA are the cause of Gaucher disease type 2 (GD2) [MIM:230900]; also known as acute neuronopathic Gaucher disease. GD2 is the most severe form and is universally progressive and fatal. It manifests soon after birth, with death generally occurring before patients reach two years of age.

Defects in GBA are the cause of Gaucher disease type 3 (GD3) [MIM:231000]; also known as subacute neuronopathic Gaucher disease. GD3 has central nervous manifestations.

Defects in GBA are the cause of Gaucher disease type 3C (GD3C) [MIM:231005]; also known as pseudo-Gaucher disease or Gaucher-like disease.

Defects in GBA are the cause of Gaucher disease perinatal lethal (GDPL) [MIM:608013]. It is a distinct form of Gaucher disease type 2, characterized by fetal onset. Hydrops fetalis, in utero fetal death and neonatal distress are prominent features. When hydrops is absent, neurologic involvement begins in the first week and leads to death within 3 months. Hepatosplenomegaly is a major sign, and is associated with ichthyosis, arthrogyrosis, and facial dysmorphism.

Note=Perinatal lethal Gaucher disease is associated with non-immune hydrops fetalis, a generalized edema of the fetus with fluid accumulation in the body cavities due to non-immune causes. Non-immune hydrops fetalis is not a diagnosis in itself but a symptom, a feature of many genetic disorders, and the end-stage of a wide variety of disorders.

Defects in GBA contribute to susceptibility to Parkinson disease (PARK) [MIM:168600]. A complex neurodegenerative disorder characterized by bradykinesia, resting tremor, muscular rigidity and postural instability. Additional features are characteristic postural abnormalities, dysautonomia, dystonic cramps, and dementia. The pathology of Parkinson disease involves the loss of dopaminergic neurons in the substantia nigra and the presence of Lewy bodies (intraneuronal accumulations of aggregated proteins), in surviving neurons in various areas of the

brain. The disease is progressive and usually manifests after the age of 50 years, although early-onset cases (before 50 years) are known. The majority of the cases are sporadic suggesting a multifactorial etiology based on environmental and genetic factors. However, some patients present with a positive family history for the disease. Familial forms of the disease usually begin at earlier ages and are associated with atypical clinical features.

Sequence similarities

Belongs to the glycosyl hydrolase 30 family.

Cellular localization

Lysosome membrane. Interaction with saposin-C promotes membrane association.

Applications

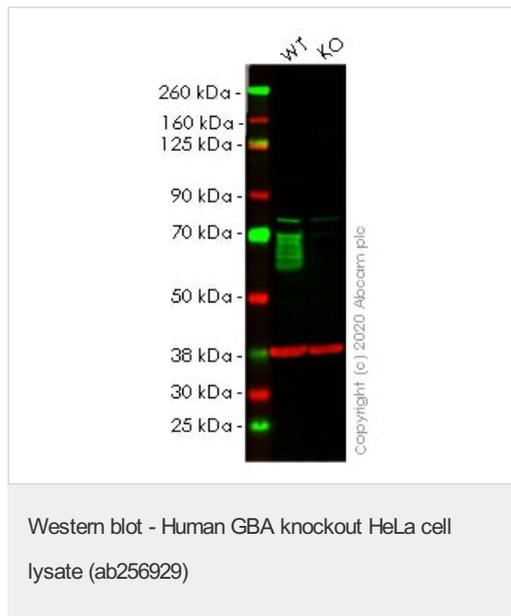
The Abpromise guarantee

Our [Abpromise guarantee](#) covers the use of ab256929 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. Predicted molecular weight: 60 kDa.

Images

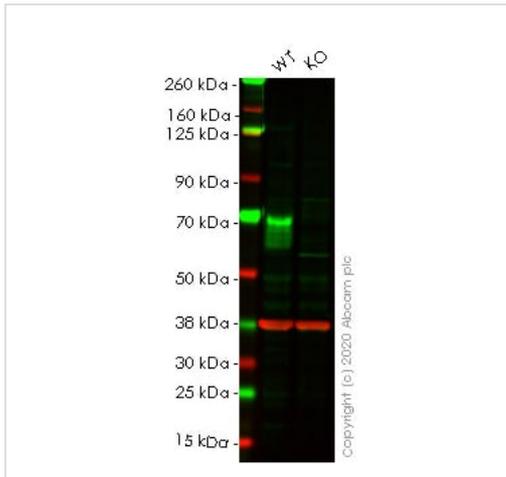


Lane 1: Wild-type HeLa cell lysate (20µg)

Lane 2: GBA knockout HeLa cell lysate (20µg)

Lanes 1- 2: Merged signal (red and green). Green - [ab128879](#) observed at 60 kDa. Red - loading control [ab8245](#) observed at 37 kDa.

[ab128879](#) Anti-GBA antibody [EPR5143(3)] was shown to specifically react with GBA in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line [ab265038](#) (knockout cell lysate ab256929) was used. Wild-type and GBA knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. [ab128879](#) and Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) 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.



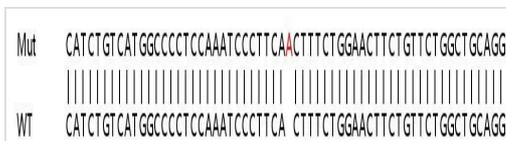
Western blot - Human GBA knockout HeLa cell lysate (ab256929)

Lane 1: Wild-type HeLa cell lysate (20µg)

Lane 2: GBA knockout HeLa cell lysate (20µg)

Lanes 1- 2: Merged signal (red and green). Green - [ab125065](#) observed at 70 kDa. Red - loading control [ab8245](#) observed at 37 kDa.

[ab125065](#) Anti-GBA antibody [EPR5142] was shown to specifically react with GBA in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line [ab265038](#) (knockout cell lysate [ab256929](#)) was used. Wild-type and GBA knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. [ab125065](#) and Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) 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.



Sanger Sequencing - Human GBA knockout HeLa cell lysate (ab256929)

Homozygous: 1 bp insertion in exon 3

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise,

please visit <https://www.abcam.com/abpromise> or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors