abcam

Product datasheet

Human MYC (c-Myc) knockout HEK-293T cell lysate ab263850

★★★★★ 1 Abreviews 画像数 8

製品の概要

製品名 Human MYC (c-Myc) knockout HEK-293T cell lysate

製品の概要

Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line HEK293T

Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, Homozygous (4N): 1bp Tinsertion (2N); 8 bp

deletion and C to T insertion (1N); 4 bp deletion in exon 2 (1N)

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.

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.

 $\label{lem:commonly} \mbox{Access thousands of knockout cell lysates, generated from commonly used cancer cell lines.}$

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 <u>limited use license</u> and <u>patent pages</u>.

アプリケーション **適用あり**: WB

1

製品の特性

保存方法

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

	1 kit
ab256769 - Human MYC knockout HEK293T cell lysate	1 x 100μg
ab255553 - Human wild-type HEK293T cell lysate	1 x 100μg

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

ターゲット情報

機能

Participates in the regulation of gene transcription. Binds DNA in a non-specific manner, yet also specifically recognizes the core sequence 5'-CAC[GA]TG-3'. Seems to activate the transcription of growth-related genes.

関連疾患

Note=Overexpression of MYC is implicated in the etiology of a variety of hematopoietic tumors. Note=A chromosomal aberration involving MYC may be a cause of a form of B-cell chronic lymphocytic leukemia. Translocation t(8;12)(q24;q22) with BTG1.

Defects in MYC are a cause of Burkitt lymphoma (BL) [MIM:113970]. A form of undifferentiated malignant lymphoma commonly manifested as a large osteolytic lesion in the jaw or as an abdominal mass. Note=Chromosomal aberrations involving MYC are usually found in Burkitt lymphoma. Translocations t(8;14), t(8;22) or t(2;8) which juxtapose MYC to one of the heavy or light chain immunoglobulin gene loci.

配列類似性

Contains 1 basic helix-loop-helix (bHLH) domain.

翻訳後修飾

Phosphorylated by PRKDC. Phosphorylation at Thr-58 and Ser-62 by GSK3 is required for

 $\label{lem:bound} \mbox{ubiquitination and degradation by the proteasome}.$

Ubiquitinated by the SCF(FBXW7) complex when phosphorylated at Thr-58 and Ser-62, leading to its degradation by the proteasome. In the nucleoplasm, ubiquitination is counteracted by USP28, which interacts with isoform 1 of FBXW7 (FBW7alpha), leading to its deubiquitination and preventing degradation. In the nucleolus, however, ubiquitination is not counteracted by USP28, due to the lack of interaction between isoform 4 of FBXW7 (FBW7gamma) and USP28, explaining the selective MYC degradation in the nucleolus. Also polyubiquitinated by the

DCX(TRUSS) complex.

細胞内局在

Nucleus > nucleoplasm. Nucleus > nucleolus.

製品の状態

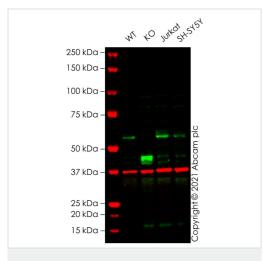
c-Myc is also expressed in the cytoplasm.

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ab263850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.

画像



Western blot - Human MYC (c-Myc) knockout HEK-293T cell lysate (ab263850)

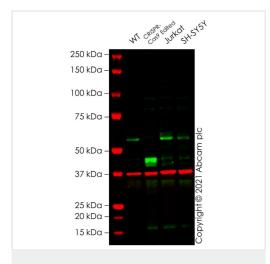
Lane 1: Wild-type HEK-293T cell lysate 20 µg

Lane 2: MYC knockout HEK-293T cell lysate 20 µg

Lane 3: Jurkat cell lysate 20 µg

Lane 4: SH-SY5Y cell lysate 20 µg

False colour image of Western blot: Anti-Myc tag antibody [Myc.A7] staining at 1/1000 dilution, shown in green; Rabbit Anti-GAPDH antibody [EPR16891] (ab181602) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab18185 was shown to bind specifically to Myc tag. A band was observed at 57 kDa in wild-type HEK-293T cell lysates with no signal observed at this size in MYC knockout cell line ab256500 (knockout cell lysate ab263850). The band observed in the knockout lysate lane below 57 kDa is likely to represent a truncated form of Myc tag. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wild-type and MYC knockout HEK-293T cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (ab216772) and Goat anti-Rabbit lgG H&L (IRDye® 680RD) preabsorbed (ab216777) at 1/20000 dilution.



Western blot - Human MYC (c-Myc) knockout HEK-293T cell lysate (ab263850)

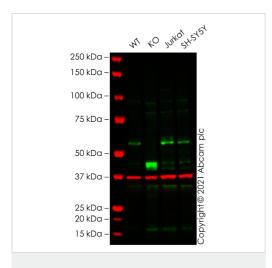
Lane 1: Wild-type HEK-293T cell lysate 20 µg

Lane 2: MYC CRISPR-Cas9 edited HEK-293T cell lysate 20 µg

Lane 3: Jurkat cell lysate 20 µg

Lane 4: SH-SY5Y cell lysate 20 µg

False colour image of Western blot: Anti-Myc tag antibody [Myc.A7] staining at 1/1000 dilution, shown in green; Rabbit Anti-GAPDH antibody [EPR16891] (ab181602) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab18185 was shown to bind specifically to Myc tag. A band was observed at 57 kDa in wild-type HEK-293T cell lysates with no signal observed at this size in MYC CRISPR-Cas9 edited cell line ab256500 (CRISPR-Cas9 edited cell lysate ab263850). The band observed in the CRISPR-Cas9 edited lysate lane below 57 kDa is likely to represent a truncated form of Myc tag. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wildtype and MYC CRISPR-Cas9 edited HEK-293T cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (ab216772) and Goat anti-Rabbit lgG H&L (IRDye® 680RD) preabsorbed (ab216777) at 1/20000 dilution.



Western blot - Human MYC (c-Myc) knockout HEK-293T cell lysate (ab263850)

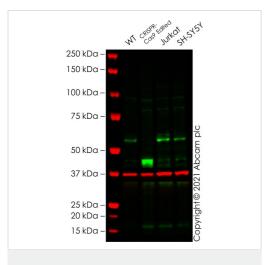
Lane 1: Wild-type HEK-293T cell lysate 20 µg

Lane 2: MYC knockout HEK-293T cell lysate 20 µg

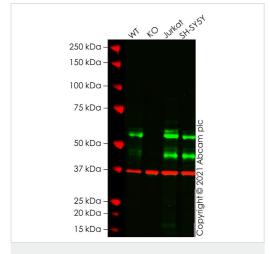
Lane 3: Jurkat cell lysate 20 µg

Lane 4: SH-SY5Y cell lysate 20 µg

False colour image of Western blot: Anti-Myc tag antibody [9E10] staining at 1/200 dilution, shown in green; Rabbit Anti-GAPDH antibody [EPR16891] (ab181602) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab32 was shown to bind specifically to Myc tag. A band was observed at 57 kDa in wild-type HEK-293T cell lysates with no signal observed at this size in MYC knockout cell line ab256500 (knockout cell lysate ab263850). The band observed in the knockout lysate lane below 57 kDa is likely to represent a truncated form of Myc tag. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wild-type and MYC knockout HEK-293T cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (ab216772) and Goat anti-Rabbit lgG H&L (IRDye® 680RD) preabsorbed (ab216777) at 1/20000 dilution.



Western blot - Human MYC (c-Myc) knockout HEK-293T cell lysate (ab263850)



Western blot - Human MYC (c-Myc) knockout HEK-293T cell lysate (ab263850)

Lane 1: Wild-type HEK-293T cell lysate 20 µg

Lane 2: MYC CRISPR-Cas9 edited HEK-293T cell lysate 20 µg

Lane 3: Jurkat cell lysate 20 µg

Lane 4: SH-SY5Y cell lysate 20 µg

False colour image of Western blot: Anti-Myc tag antibody [9E10] staining at 1/200 dilution, shown in green; Rabbit Anti-GAPDH antibody [EPR16891] (ab181602) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab32 was shown to bind specifically to Myc tag. A band was observed at 57 kDa in wild-type HEK-293T cell lysates with no signal observed at this size in MYC CRISPR-Cas9 edited cell line ab256500 (CRISPR-Cas9 edited cell lysate ab263850). The band observed in the CRISPR-Cas9 edited lysate lane below 57 kDa is likely to represent a truncated form of Myc tag. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wild-type and MYC CRISPR-Cas9 edited HEK-293T cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (ab216772) and Goat anti-Rabbit lgG H&L (IRDye® 680RD) preabsorbed (ab216777) at 1/20000 dilution.

Lane 1: Wild-type HEK-293T cell lysate 20 µg

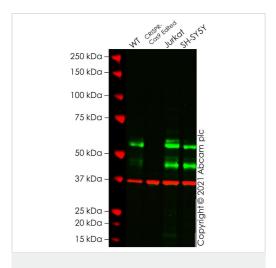
Lane 2: MYC knockout HEK-293T cell lysate 20 µg

Lane 3: Jurkat cell lysate 20 µg

Lane 4: SH-SY5Y cell lysate 20 µg

False colour image of Western blot: Anti-c-Myc antibody [Y69] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab32072 was shown to bind specifically to c-Myc. A band was observed at 45/57 kDa in wild-type HEK-293T cell lysates with no signal observed at this size in MYC knockout cell line ab256500 (knockout cell lysate ab263850). The band observed in the knockout lysate lane below 45/57 kDa is likely to represent a truncated form of c-Myc. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wild-type and MYC knockout HEK-293T cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a

nitrocellulose membrane. Membranes were blocked in 5 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed (ab216776) at 1/20000 dilution.



Western blot - Human MYC (c-Myc) knockout HEK-293T cell lysate (ab263850)

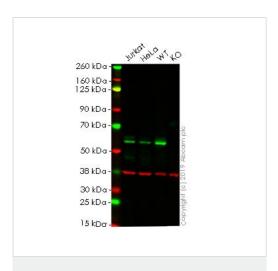
Lane 1: Wild-type HEK-293T cell lysate 20 µg

Lane 2: MYC CRISPR-Cas9 edited HEK-293T cell lysate 20 µg

Lane 3: Jurkat cell lysate 20 µg

Lane 4: SH-SY5Y cell lysate 20 µg

False colour image of Western blot: Anti-c-Myc antibody [Y69] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab32072 was shown to bind specifically to c-Myc. A band was observed at 45/57 kDa in wildtype HEK-293T cell lysates with no signal observed at this size in MYC CRISPR-Cas9 edited cell line ab256500 (CRISPR-Cas9 edited cell lysate ab263850). The band observed in the CRISPR-Cas9 edited lysate lane below 45/57 kDa is likely to represent a truncated form of c-Myc. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wild-type and MYC CRISPR-Cas9 edited HEK-293T cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 5 % milk in TBS-0.1 % Tween[®] 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) at 1/20000 dilution.



Western blot - Human MYC knockout HEK293T cell lysate (ab263850)

Lane 1: Jurkat cell lysate (20 µg)

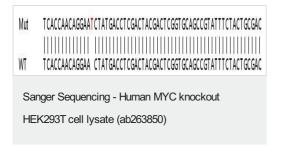
Lane 2: HeLa cell lysate (20 µg)

Lane 3: Wild-type HEK-293T cell lysate (20 µg)

Lane 4: MYC knockout HEK-293T cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - <u>ab32072</u> observed at 57 kDa. Red - loading control, <u>ab8245</u> observed at 37 kDa.

ab32072 was shown to react with MYC in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line ab256500 (knockout cell lysate ab263850) was used. Wild-type and MYC knockout samples were subjected to SDS-PAGE. ab32072 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.



Homozygous: 1 bp insertion in exon 2

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.co.jp/abpromise or contact our technical team.

Terms and conditions

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