

Human ATG3 knockout HEK-293T cell lysate ab257363

画像数 3

製品の概要

製品名	Human ATG3 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: 1 bp deletion in exon 1.
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.

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

製品の特性

保存方法

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

内容	1 kit
ab260972 - Human ATG3 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

E2-like enzyme involved in autophagy and mitochondrial homeostasis. Catalyzes the conjugation of ATG8-like proteins (GABARAP, GABARAPL1, GABARAPL2 or MAP1LC3A) to phosphatidylethanolamine (PE). PE-conjugation to ATG8-like proteins is essential for autophagy. Preferred substrate is MAP1LC3A. Also acts as an autocatalytic E2-like enzyme, catalyzing the conjugation of ATG12 to itself, ATG12 conjugation to ATG3 playing a role in mitochondrial homeostasis but not in autophagy. ATG7 (E1-like enzyme) facilitates this reaction by forming an E1-E2 complex with ATG3.

組織特異性

Widely expressed, with a highest expression in heart, skeletal muscle, kidney, liver and placenta.

配列類似性

Belongs to the ATG3 family.

翻訳後修飾

Conjugated to ATG12 at Lys-243. ATG12-conjugation plays a role in regulation of mitochondrial homeostasis and cell death, while it is not involved in PE-conjugation to ATG8-like proteins and autophagy.

細胞内局在

Cytoplasm.

アプリケーション

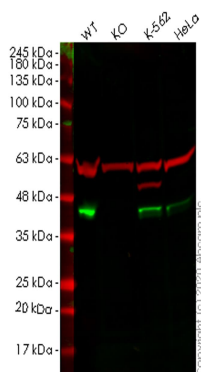
The Abpromise guarantee

Abpromise保証は、次のテスト済みアプリケーションにおけるab257363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: 35 kDa.

画像



Western blot - Human ATG3 knockout HEK293T cell lysate (ab257363)

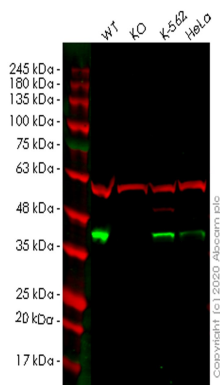
Lane 1: Wild-type HEK293T cell lysate (20 ug)

Lane 2: ATG3 knockout HEK293T cell lysate (20 ug)

Lane 3: K-562 cell lysate (20 ug)

Lane 4: HeLa cell lysate (20 ug)

ab108251 was shown to specifically react with ATG3 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line **ab266707** (knockout cell lysate ab257363) was used. Wild-type and ATG3 knockout samples were subjected to SDS-PAGE. **ab108251** and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) 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 ATG3 knockout HEK293T cell lysate (ab257363)

Lane 1: Wild-type HEK293T cell lysate (20 ug)

Lane 2: ATG3 knockout HEK293T cell lysate (20 ug)

Lane 3: K-562 cell lysate (20 ug)

Lane 4: HeLa cell lysate (20 ug)

ab108282 was shown to specifically react with ATG3 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line **ab266707** (knockout cell lysate ab257363) was used. Wild-type and ATG3 knockout samples were subjected to SDS-PAGE. **ab108282** and Anti-alpha Tubulin antibody [DM1A] - Loading Control? (**ab7291**) 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.

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Mut  GAAAGGCACTGGAAGTGGCTGAGTACCTGACCC - GGTCTCAAGGTAAGCCAGGCCAGGG
      |||||||||||||||||||||||||||||||||||
WT   GAAAGGCACTGGAAGTGGCTGAGTACCTGACCCCGGTCTCAAGGTAAGCCAGGCCAGGG
  
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Sanger Sequencing - Human ATG3 knockout HEK293T cell lysate (ab257363)

Homozygous: 1 bp deletion in exon 1

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