abcam

Product datasheet

Mouse CD68 knockout RAW 264.7 cell line ab280047

画像数 3

製品の概要

製品名	Mouse CD68 knockout RAW 264.7 cell line
Parental Cell Line	RAW 264.7
Organism	Mouse
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
アプリケーション	**適用あり:** WB
Biosafety level	2
特記事項	**Recommended control:** Mouse wild-type RAW 264.7 cell line (**ab275474**). Please note a wild-type cell line is not automatically included with a knockout cell line order, if required please add recommended wild-type cell line at no additional cost using the code WILDTYPE-TMTK1. **Cryopreservation cell medium:** Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose. **Culture medium:** DMEM + 10% FBS **Initial handling guidelines:** Upon arrival, the vial should be stored in liquid nitrogen vapor phase and not at -80°C. Storage at -80°C may result in loss of viability. 1. Thaw the vial in 37°C water for bath approximately 1-2 minutes. 2. Transfer the cell suspension (0.8 mL) to a 15 mL/50 mL conical sterile polypropylene centrifuge tube containing 8.4 mL pre-warmed culture medium, wash vial with an additional 0.8 mL culture medium (total volume 10 mL) to collect remaining cells, and centrifuge at 201 x g (rcf) for 5 minutes at room temperature. 10 mL represents minimum recommended dilution. 20 mL represents maximum recommended dilution. 3. Resuspend the cell pellet in 5 mL pre-warmed culture medium and count using a haemocytometer or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of $2x10^5$ cells/mL. Seeding density is given as a guide only and should be scaled to align with individual lab schedules. 4. Incubate the culture at 37°C incubator with 5% CO_2. Cultures should be monitored daily. **Subculture guidelines:** All seeding densities should be based on cell counts gained by established methods. A guide seeding density of $2x10^5$ cells/mL is recommended. This product is subject to limited use licenses from The Broad Institute and ERS Genomics 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**.

We will provide viable cells that proliferate on revival.

製品の特性

Number of cells	1 x 10^6 cells/vial, 1 mL
Adherent /Suspension	Adherent
Tissue	Lymphatic
Cell type	leukaemic monocyte macrophage
Disease	Carcinoma
Gender	Male
Mycoplasma free	Yes
保存方法	Shipped on Dry Ice. Store in liquid nitrogen.
バッファー	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

ターゲット情報

機能	Could play a role in phagocytic activities of tissue macrophages, both in intracellular lysosomal metabolism and extracellular cell-cell and cell-pathogen interactions. Binds to tissue- and organ-specific lectins or selectins, allowing homing of macrophage subsets to particular sites. Rapid recirculation of CD68 from endosomes and lysosomes to the plasma membrane may allow macrophages to crawl over selectin-bearing substrates or other cells.
組織特異性	Highly expressed by blood monocytes and tissue macrophages. Also expressed in lymphocytes, fibroblasts and endothelial cells. Expressed in many tumor cell lines which could allow them to attach to selectins on vascular endothelium, facilitating their dissemination to secondary sites.
配列類似性	Belongs to the LAMP family.
翻訳後修飾	N- and O-glycosylated.
細胞内局在	Cell membrane and Endosome membrane. Lysosome membrane.

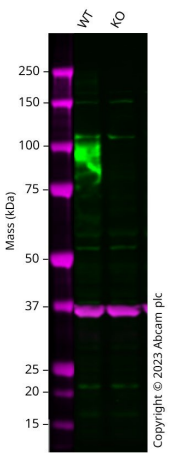
アプリケーション

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ab280047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 - Mouse CD68 knockout RAW 264.7 cell line (ab280047)

All lanes : Anti-CD68 antibody [RM1031] (**ab303565**) at 1/1000 dilution

Lane 1 : Wild-type RAW 264.7 cell lysate
Lane 2 : CD68 knockout RAW 264.7 cell lysate

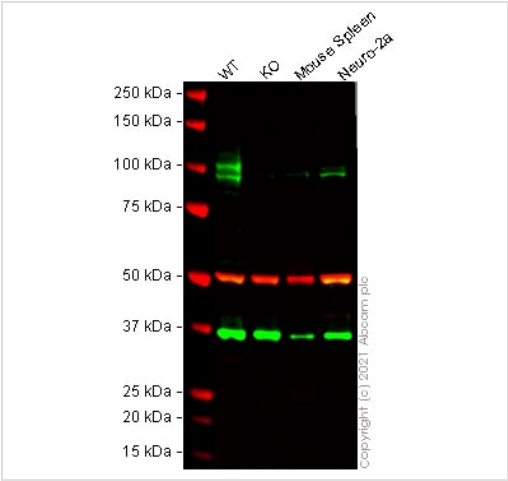
Lysates/proteins at 20 µg per lane.

Developed using the ECL technique.

Performed under reducing conditions.

Observed band size: 100 kDa

Western blot: Anti-CD68 antibody [RM1031] (**ab303565**) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (**ab8245**) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, **ab303565** was shown to bind specifically to CD68. A band was observed at 100 kDa in wild-type RAW 264.7 cell lysates with no signal observed at this size in CD68 knockout cell line. To generate this image, wild-type and CD68 knockout RAW 264.7 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 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.



Western blot - Mouse CD68 knockout RAW 264.7 cell line (ab280047)

All lanes : Anti-CD68 antibody (**ab125212**) at 0.2 µg/ml

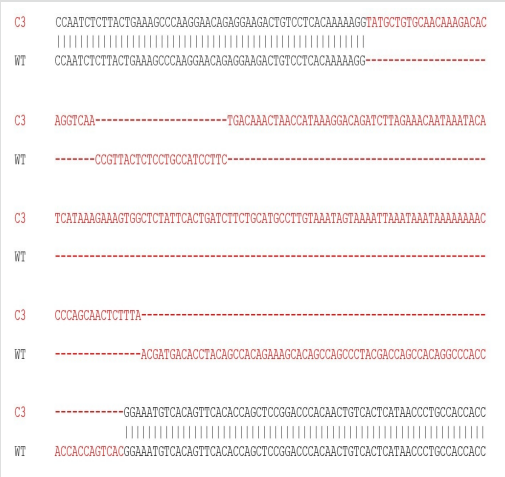
Lane 1 : Wild-type RAW 264.7 cell lysate
Lane 2 : CD68 knockout RAW 264.7 cell lysate
Lane 3 : Mouse spleen cell lysate
Lane 4 : Neuro-2a cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 95-102 kDa

False colour image of Western blot: Anti-CD68 antibody staining at 0.2 µg/ml, shown in green; Mouse anti-Alpha Tubulin [DM1A] (**ab7291**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab125212** was shown to bind specifically to CD68. A band was observed at 95-102 kDa in wild-type RAW 264.7 cell lysates with no signal observed at this size in CD68 knockout cell line ab280047 (knockout cell lysate **ab280106**). To generate this image, wild-type and CD68 knockout RAW 264.7 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution 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.



Sanger Sequencing - Mouse CD68 knockout RAW 264.7 cell line (ab280047)

Mouse Cd68 KO in Raw264.7 Cells with 28 bp insertion-13 bp deletion-135 bp insertion-72 bp deletion in Exon 2.

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