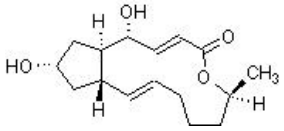
abcam

Product datasheet

# Brefeldin A, Inhibitor of ADP-ribosylation factor ab120299

★★★★★ 1 Abreviews 6 References 画像数 10

## 製品の概要

| | |
|---|---|
| **製品名** | Brefeldin A, Inhibitor of ADP-ribosylation factor |
| **製品の詳細** | Inhibitor of ADP-ribosylation factor |
| **生理活性の詳細** | Reversible inhibitor of protein translocation from the endoplasmic reticulum to the Golgi apparatus. Inhibits binding of the cytosolic coat protein, β-COP and ADP-ribosylation factor (ARF) to Golgi membranes and inhibits GDP-GTP exchange. |
| **CAS 番号** | 20350-15-6 |
| **構造式** |  |

## 製品の特性

| | |
|---|---|
| **体系名** | (1*R*,2*E*,6*S*,10*E*,11a*S*,13*S*,14a*R*)-1,13-Dihydroxy-6-methyl-1,6,7,8,9,11a,12,13,14,14a-decahydro-4*H*-cyclopenta[*f*]oxacyclotridecin-4-one |
| **分子量** | 280.36 |
| **分子式** | $C_{16}H_{24}O_4$ |
| **PubChem 登録番号** | 5287620 |
| **保存方法** | Store at -20°C. Store under desiccating conditions. The product can be stored for up to 12 months. |
| **溶解性** | Soluble in DMSO to 50 mM |
| **使用に関する注意** | Wherever possible, you should prepare and use solutions on the same day. However, if you need to make up stock solutions in advance, we recommend that you store the solution as aliquots in tightly sealed vials at -20°C. Generally, these will be useable for up to one month. Before use, and prior to opening the vial we recommend that you allow your product to equilibrate to room temperature for at least 1 hour.<br>Toxic, refer to SDS for further information.<br>Need more advice on solubility, usage and handling? Please visit our **frequently asked questions (FAQ) page** for more details. |
| **SMILES 線形表記** | O[C@@H]1C[C@H]2[C@H](O)C=CC(=O)O[C@@H](C)CCCC=C[C@@H]2C1 |

**由来** *Eupenicillium brefeldianum*

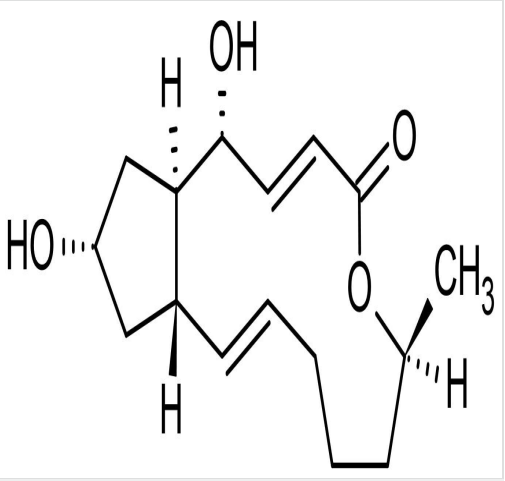
## アプリケーション

**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ab120299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。

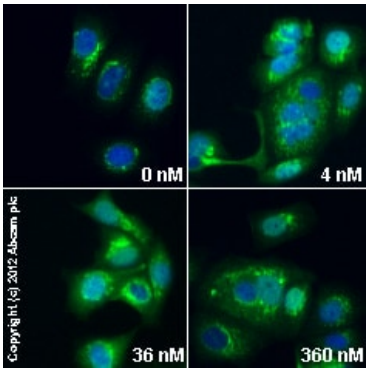
| アプリケーション | Abreviews | 特記事項 |
|---|---|---|
| **Functional Studies** | | Use at an assay dependent concentration. |

## 画像




Chemical Structure - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

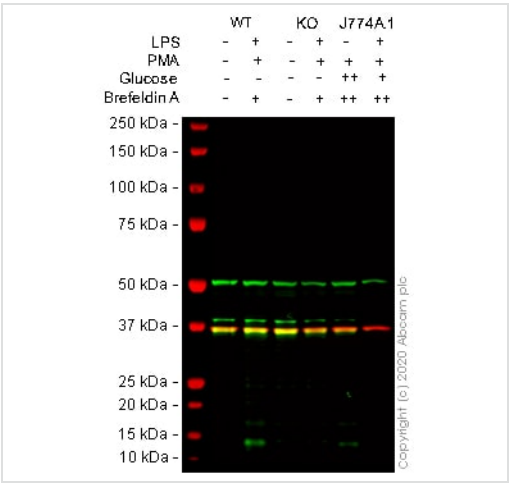
2D chemical structure image of ab120299, Brefeldin A, Inhibitor of ADP-ribosylation factor




Immunocytochemistry/ Immunofluorescence - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

**ab84340** staining golgin-97 in MCF7 cells treated with brefeldin A (ab120299), by ICC/IF. Increase in Golgin-97 expression correlates with increased concentration of brefeldin A, as described in literature.

The cells were incubated at 37°C for 1.5 h in media containing different concentrations of ab120299 (brefeldin A) in DMSO, fixed with 4% formaldehyde for 10 minutes at room temperature and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with **ab84340** (5 µg/ml) was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A DyLight 488 goat anti-rabbit polyclonal antibody (**ab96899**) at 1/250 dilution was used as the secondary antibody. Nuclei were counterstained with DAPI and are shown in blue.




Western blot - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

**All lanes :** Anti-MCP3 antibody [EPR22649-155] (**ab228979**) at 1/1000 dilution

**Lane 1 :** Wild-type RAW 264.7 untreated control cell lysate
**Lane 2 :** Wild-type RAW 264.7 PMA treated (80 nM, 24 h) plus LPS treated (100 ng/ml, 6 h) and Brefeldin A (ab120299) treated (5 µg/ml, 5 h) cell lysate
**Lane 3 :** CCL7/MCP3 knockout RAW 264.7 untreated cell lysate
**Lane 4 :** CCL7/MCP3 knockout RAW 264.7 PMA treated (80 nM, 24 h) plus LPS treated (100 ng/ml, 6 h) and Brefeldin A (ab120299) treated (5 µg/ml, 5 h) cell lysate
**Lane 5 :** J774A.1 Glucose treated (138.8 mMol/L, 8 h) plus Brefeldin A treated (5 µg/ml, 6 h) and Brefeldin A (ab120299) treated (5 µg/ml, 5 h) cell lysate
**Lane 6 :** J774A.1 Glucose treated (5.6 mMol/L, 8 h) plus Brefeldin A treated (5 µg/ml, 6 h) and Brefeldin A (ab120299) treated (5 µg/ml, 5 h) cell lysate

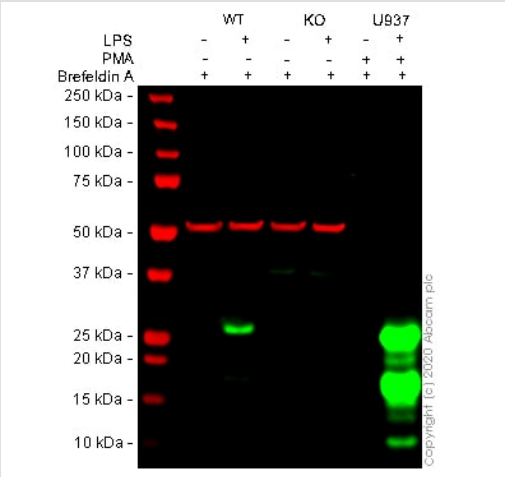
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

**Observed band size:** 14 kDa

**Lanes 1 - 6:** Merged signal (red and green). Green - **ab228979** observed at 14 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

**ab228979** was shown to react with MCP3 in RAW 264.7 wild-type cells in Western blot with loss of signal observed in CCL7 knockout sample. Wild-type and CCL7 knockout RAW 264.7 cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab228979** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 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 (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.




Western blot - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

**All lanes :** Anti-TNF alpha antibody [EPR22598-212] (**ab255275**) at 1/1000 dilution

**Lane 1 :** Wild-type THP-1 Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 2 :** Wild-type THP-1 LPS treated (100 ng/ml, 16 h) and Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 3 :** TNF alpha knockout THP-1 Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 4 :** TNF alpha knockout THP-1 LPS treated (100 ng/ml, 16 h) and Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 5 :** U937 PMA treated (10 mM, 2 days) plus 16 h no treatment and Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 6 :** U937 PMA treated (10 mM, 2 days) and LPS treated (1 µg/ml, 16 h) plus Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate

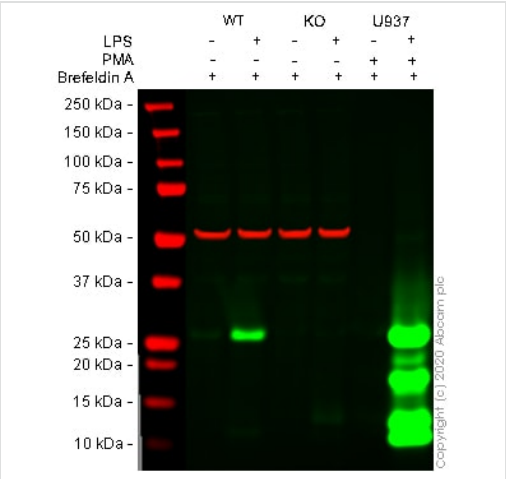
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

**Observed band size:** 26 kDa

**Lanes 1 - 6:** Merged signal (red and green). Green - **ab255275** observed at 26 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.

**ab255275** was shown to react with TNF alpha in THP-1 wild-type cells in Western blot with loss of signal observed in TNF knockout sample. Wild-type and TNF knockout THP-1 cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab255275** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 1000 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 (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.




Western blot - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

**All lanes :** Anti-TNF alpha antibody [EPR19147] (**ab183218**) at 1/1000 dilution

**Lane 1 :** Wild-type THP-1 Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 2 :** Wild-type THP-1 LPS treated (100 ng/ml, 16 h) and Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 3 :** TNF alpha knockout THP-1 Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 4 :** TNF alpha knockout THP-1 LPS treated (100 ng/ml, 16 h) and Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 5 :** U937 PMA treated (10 mM, 2 days) plus 16 h no treatment and Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate
**Lane 6 :** U937 PMA treated (10 mM, 2 days) and LPS treated (1 µg/ml, 16 h) plus Brefeldin A (ab120299) treated (5 µg/ml, 4 h) cell lysate

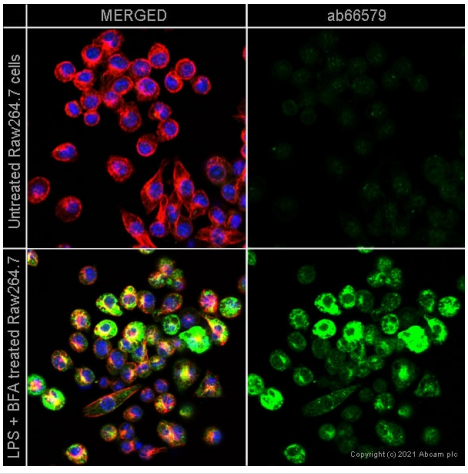
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

**Observed band size:** 26 kDa

**Lanes 1 - 6:** Merged signal (red and green). Green - **ab183218** observed at 26 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.

**ab183218** was shown to react with TNF alpha in THP-1 wild-type cells in Western blot with loss of signal observed in TNF knockout sample. Wild-type and TNF knockout THP-1 cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab183218** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 1000 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 (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.

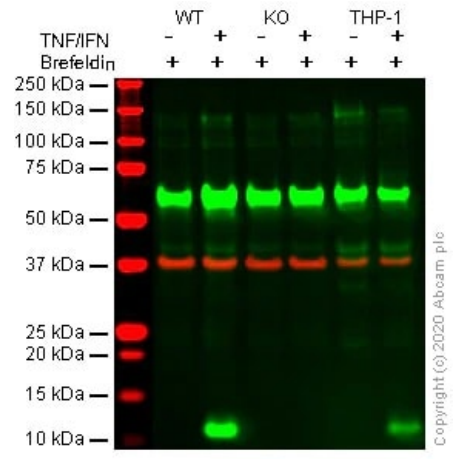



Immunocytochemistry - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

**ab66579** staining TNF alpha in Raw264.7 cells. The cells were treated with LPS for 7 hours and Brefeldin A (ab120299) for the final 3 hours. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at 4°C with **ab66579** at 1µg/ml and **ab7291**, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with **ab150081**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and **ab150120**, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Also suitable in cells fixed with 4% paraformaldehyde (10 min).

Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.




Western blot - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

**All lanes :** Anti-IP10 antibody [EPR20764] (**ab214668**) at 1/1000 dilution

**Lane 1 :** Wild-type A549 Brefeldin A (ab120299)-treated (5ug/ml, 6h) cell lysate
**Lane 2 :** Wild-type A549 IFN-y (**ab259377**) (100 ng/ml, 32 h) and TNF-alpha (**ab259410**) (10 ng/ml, 32h), and Brefeldin A (ab120299)-treated (5ug/ml for the last 6h) cell lysate
**Lane 3 :** IP10 knockout A549 Brefeldin A (ab120299)-treated (5ug/ml, 6h) cell lysate
**Lane 4 :** IP10 knockout A549 IFN-y (**ab259377**) (100ng/ml, 32h) and TNF-alpha (**ab259410**) (10ng/ml, 32h), and Brefeldin A (ab120299)-treated (5ug/ml for the last 6h) cell lysate
**Lane 5 :** THP-1 Brefeldin A (ab120299)-treated (5ug/ml, 6h) cell lysate
**Lane 6 :** THP-1 IFN-y (**ab259377**) (200ng/ml, 24h) and LPS (50ng/ml, 24h)-treated for 24 hours, and Brefeldin A (ab120299)-treated (5ug/ml for the last 6h) cell lysate

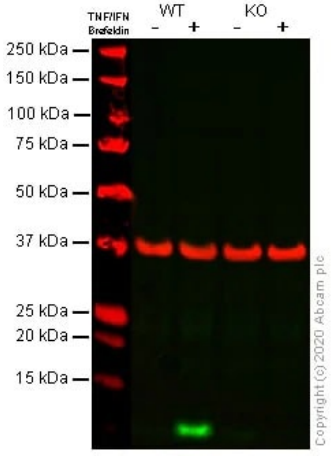
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

**Observed band size:** 11 kDa

**Lanes 1 - 6:** Merged signal (red and green). Green - **ab214668** observed at 11 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

**ab214668** was shown to react with IP10 in wild-type A549 cells in western blot with loss of signal observed in IP10 knockout cell line **ab266971** (knockout cell lysate **ab256888**). Wild-type and IP10 knockout A549 cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab214668** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 1000 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® 800CW) preabsorbed (**ab216772**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.




Western blot - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

**All lanes :** Anti-IP10 antibody [EPR7850] (**ab137018**) at 1/500 dilution

**Lane 1 :** Wild-type A549 Brefeldin A (ab120299)-treated (5ug/ml, 6h) cell lysate
**Lane 2 :** Wild-type A549 IFN-y (**ab259377**) (100 ng/ml, 32 h) and TNF-alpha (**ab259410**) (10 ng/ml) for 32 hours, and Brefeldin A (ab120299)-treated (5ug/ml for the last 6h) cell lysate
**Lane 3 :** IP10 knockout A549 Brefeldin A (ab120299)-treated (5ug/ml, 6h) cell lysate
**Lane 4 :** IP10 knockout A549 IFN-y (**ab259377**) (100 ng/ml, 32 h) and TNF-alpha (**ab259410**) (10 ng/ml) for 32 hours, and Brefeldin A (ab120299)-treated (5ug/ml for the last 6h) cell lysate

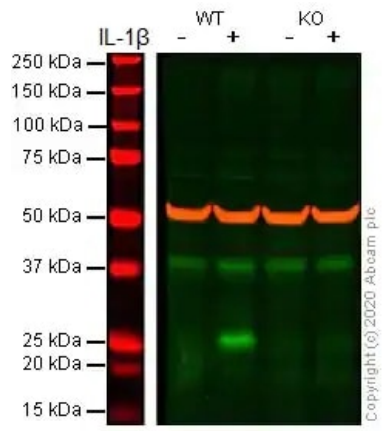
Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

**Observed band size:** 11 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - **ab137018** observed at 11 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

**ab137018** was shown to react with IP10 in A549 wild-type cells in western blot with loss of signal observed in IP10 knockout cell line **ab266969** (IP10 knockout cell lysate **ab256886**). A549 wild-type and IP10 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab137018** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 500 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® 800CW) preabsorbed (**ab216772**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.




Western blot - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

**All lanes :** Anti-IL-6 antibody [EPR21711] (**ab233706**) at 1/1000 dilution

**Lane 1 :** Wild-type A549 Brefeldin A (ab120299)-treated (5ug/ml, 4h) cell lysate
**Lane 2 :** Wild-type A549 IL-1ß (**ab259387**) (20 ng/ml, 24h) and Brefeldin A (ab120299)-treated (5 ug/ml for the last 4h) cell lysate
**Lane 3 :** IL-6 knockout A549 Brefeldin A (ab120299)-treated (5ug/ml, 4h) cell lysate
**Lane 4 :** IL-6 knockout A549 IL-1ß (**ab259387**) (20 ng/ml, 24h) and Brefeldin A (ab120299)-treated (5 ug/ml for the last 4h) cell lysate

Lysates/proteins at 30 µg per lane.

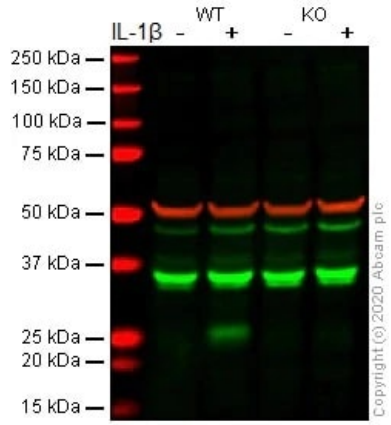
Performed under reducing conditions.

**Observed band size:** 25 kDa
**Additional bands at:** 40 kDa (possible non-specific binding)

**Lanes 1 - 4:** Merged signal (red and green). Green - **ab233706** observed at 25 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

**ab233706** was shown to react with IL-6 in wild-type A549 cells in western blot with loss of signal observed in IL-6 knockout cell line **ab273751** (knockout cell lysate **ab275501**). Wild-type and IL-6 knockout A549 cell lysates were subjected to SDS-PAGE.

Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab233706** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A] overnight at 4°C at a 1 in 1000 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 (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.




Western blot - Brefeldin A, Inhibitor of ADP-ribosylation factor (ab120299)

**All lanes :** Anti-IL-6 antibody [EPR20653] (**ab214429**) at 1/1000 dilution

**Lane 1 :** Wild-type A549 Brefeldin A (ab120299)-treated (5ug/ml, 4h) cell lysate
**Lane 2 :** Wild-type A549 IL-1ß (**ab259387**) (20 ng/ml, 24h) and Brefeldin A (ab120299)-treated (5 ug/ml for the last 4h) cell lysate
**Lane 3 :** IL-6 knockout A549 Brefeldin A (ab120299)-treated (5ug/ml, 4h) cell lysate
**Lane 4 :** IL-6 knockout A549 IL-1ß (**ab259387**) (20 ng/ml, 24h) and Brefeldin A (ab120299)-treated (5 ug/ml for the last 4h) cell lysate

Lysates/proteins at 30 µg per lane.

Performed under reducing conditions.

**Lanes 1 - 4:** Merged signal (red and green). Green - **ab214429** observed at 25 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A] observed at 55kDa.

**ab214429** was shown to react with IL-6 in wild-type A549 cells in western blot with loss of signal observed in IL-6 knockout cell line **ab273751** (knockout cell lysate **ab275501**). Wild-type A549 and IL-6 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab214429** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A] overnight at 4°C at a 1 in 1000 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 (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

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