# abcam

## Product datasheet

## Anti-IL-6 antibody [EPR20653] ab214429



ייבעדיו RabMAb

#### 画像数5 4 References

#### 製品の概要

製品名 Anti-IL-6 antibody [EPR20653]

製品の詳細 Rabbit monoclonal [EPR20653] to IL-6

由来種 Rabbit

アプリケーション 適用あり: WB, ICC/IF, IP

種交差性 交差種: Human

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: HUVEC treated with 0.5 µg/ml Lipopolysaccharides (LPS) for 24 hours and 300 ng/ml

> Brefeldin A (BFA) for 20 hours whole cell lysate; Wild-type A549 IL-1ß (ab259387) (20 ng/ml, 24h) and Brefeldin A (ab120299)-treated (5 ug/ml for the last 4h) cell lysate ICC/IF: HUVEC treated with Lipopolysaccharides (LPS) (0.5 µg/ml 24h) and Brefeldin A (BFA) (300 ng/ml 20h) cells. IP: HUVEC treated with 0.5 µg/ml Lipopolysaccharides (LPS) for 24 hours and 300 ng/ml

Brefeldin A (BFA) for 20 hours whole cell lysate.

特記事項 This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

#### 製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

バッファー pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 0.05% BSA, 40% Glycerol (glycerin, glycerine), PBS

精製度 Protein A purified

**ポリ**(モノ モノクローナル **ウローン名** EPR20653

アイソタイプ IgG

#### 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ab214429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		1/1000. Detects a band of approximately 21-28 kDa (predicted molecular weight: 23 kDa).
ICC/IF		1/500.
IP		1/30.

#### ターゲット情報

機能 Cytokine with a wide variety of biological functions. It is a potent inducer of the acute phase

response. Plays an essential role in the final differentiation of B-cells into Ig-secreting cells Involved in lymphocyte and monocyte differentiation. It induces myeloma and plasmacytoma growth and induces nerve cells differentiation Acts on B-cells, T-cells, hepatocytes, hematopoeitic

progenitor cells and cells of the CNS. Also acts as a myokine. It is discharged into the

bloodstream after muscle contraction and acts to increase the breakdown of fats and to improve

insulin resistance.

関連疾患 Genetic variations in IL6 are associated with susceptibility to rheumatoid arthritis systemic

juvenile (RASJ) [MIM:604302]. An inflammatory articular disorder with systemic-onset beginning

before the age of 16. It represents a subgroup of juvenile arthritis associated with severe

extraarticular features and occasionally fatal complications. During active phases of the disorder, patients display a typical daily spiking fever, an evanescent macular rash, lymphadenopathy,

hepatosplenomegaly, serositis, myalgia and arthritis.

Note=A IL6 promoter polymorphism is associated with a lifetime risk of development of Kaposi

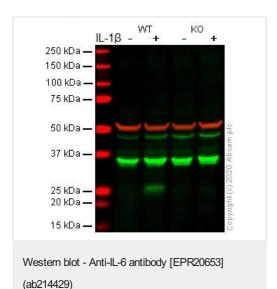
sarcoma in HIV-infected men.

配列類似性 Belongs to the IL-6 superfamily.

翻訳後修飾 N- and O-glycosylated.

細胞内局在 Secreted.

## 画像



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

**Lane 1 :** Wild-type A549 Brefeldin A (<u>ab120299</u>)-treated (5ug/ml, 4h) cell lysate

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

Lysates/proteins at 30 µg per lane.

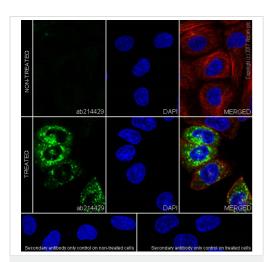
Performed under reducing conditions.

**Predicted band size:** 23 kDa **Observed band size:** 25 kDa

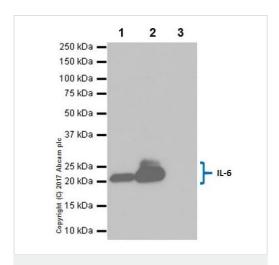
Additional bands at: 35 kDa (possible non-specific binding)

**Lanes 1 - 4:** Merged signal (red and green). Green - ab214429 observed at 25 kDa. Red - loading control <u>ab7291</u> (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 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 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.



Immunocytochemistry/ Immunofluorescence - Anti-IL-6 antibody [EPR20653] (ab214429)



Immunoprecipitation - Anti-IL-6 antibody [EPR20653] (ab214429)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HUVEC (human umbilical vein endothelial cell line) cells, untreated or treated with Lipopolysaccharides (LPS) (0.5 µg/ml 24 hours) and Brefeldin A (BFA) (300 ng/ml 20 hours), labeling IL6 with ab214429 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing the cytoplasmic expression increased on HUVEC cells treated with Lipopolysaccharides (LPS) (0.5 µg/ml, 24 hours) and Brefeldin A (BFA) (300 ng/ml, 20 hours).

The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) (ab195889) (red) at 1/200 dilution.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution.

IL6 was immunoprecipitated from 0.35 mg of HUVEC (human umbilical vein endothelial cell line) treated with Lipopolysaccharides (LPS) and Brefeldin A (BFA) whole cell lysate with ab214429 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab214429 at 1/1,000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10.000 dilution

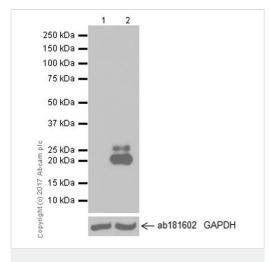
Lane 1: HUVEC treated with 0.5  $\mu$ g/ml LPS for 24 hours and 300 ng/ml BFA for 20 hours whole cell lysate 10  $\mu$ g (Input).

Lane 2: ab214429 IP in HUVEC treated with 0.5  $\mu$ g/ml LPS for 24 hours and 300 ng/ml BFA for 20 hours whole cell lysate (+).

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab214429 in HUVEC treated with 0.5  $\mu$ g/ml LPS for 24 hours and 300 ng/ml BFA for 20 hours whole cell lysate (-).

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.



Western blot - Anti-IL-6 antibody [EPR20653] (ab214429)

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

**Lane 1 :** Untreated HUVEC (human umbilical vein endothelial cell line) whole cell lysate

**Lane 2 :** HUVEC treated with 0.5  $\mu$ g/ml Lipopolysaccharides (LPS) for 24 hours and 300 ng/ml Brefeldin A (BFA) for 20 hours, whole cell lysate

Lysates/proteins at 10 µg per lane.

### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Developed using the ECL technique.

Predicted band size: 23 kDa

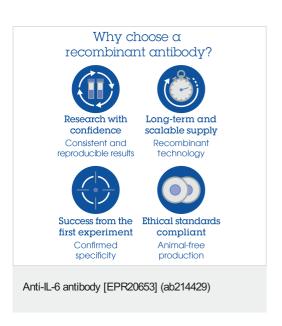
Observed band size: 21-28 kDa

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

The MW observed is consistent with the literature (PMID:2523818,

PMID: 2783321).



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