abcam

ab233551

Anti-IL-6 antibody [EPR22565-204]

Recombinant | RabMAb | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR22565-204
Purification technique	Affinity purification Protein A
Concentration	0.395 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	1/20
Notes	-

WB

Tested

Species	Human
Dilution info	1/1000
Notes	-

IHC-P

Not recommended

Species	Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/100
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/400
Notes	-

Target data

See full target information IL6

Function Cytokine with a wide variety of biological functions in immunity, tissue regeneration, and metabolism. Binds to IL6R, then the complex associates to the signaling subunit IL6ST/gp130 to trigger the intracellular IL6-signaling pathway (Probable). The interaction with the membrane-bound IL6R and IL6ST stimulates

'classic signaling', whereas the binding of IL6 and soluble IL6R to IL6ST stimulates 'trans-signaling'. Alternatively, 'cluster signaling' occurs when membrane-bound IL6:IL6R complexes on transmitter cells activate IL6ST receptors on neighboring receiver cells (Probable). IL6 is a potent inducer of the acute phase response. Rapid production of IL6 contributes to host defense during infection and tissue injury, but excessive IL6 synthesis is involved in disease pathology. In the innate immune response, is synthesized by myeloid cells, such as macrophages and dendritic cells, upon recognition of pathogens through toll-like receptors (TLRs) at the site of infection or tissue injury (Probable). In the adaptive immune response, is required for the differentiation of B cells into immunoglobulin-secreting cells. Plays a major role in the differentiation of CD4(+) T cell subsets. Essential factor for the development of T follicular helper (Tfh) cells that are required for the induction of germinal-center formation. Required to drive naive CD4(+) T cells to the Th17 lineage. Also required for proliferation of myeloma cells and the survival of plasmablast cells (By similarity). Acts as an essential factor in bone homeostasis and on vessels directly or indirectly by induction of VEGF, resulting in increased angiogenesis activity and vascular permeability (PubMed:12794819, PubMed:17075861). Induces, through 'trans-signaling' and synergistically with IL1B and TNF, the production of VEGF (PubMed:12794819). Involved in metabolic controls, is discharged into the bloodstream after muscle contraction increasing lipolysis and improving insulin resistance (PubMed:20823453). 'Trans-signaling' in central nervous system also regulates energy and glucose homeostasis (By similarity). Mediates, through GLP-1, crosstalk between insulin-sensitive tissues, intestinal L cells and pancreatic islets to adapt to changes in insulin demand (By similarity). Also acts as a myokine (Probable). Plays a protective role during liver injury, being required for maintenance of tissue regeneration (By similarity). Also has a pivotal role in iron metabolism by regulating HAMP/hepcidin expression upon inflammation or bacterial infection (PubMed:15124018). Through activation of IL6ST-YAP-NOTCH pathway, induces inflammation-induced epithelial regeneration (By similarity).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

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

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	Interleukin-6 (IL-6) a cytokine also known as IFN-beta2 plays a significant role in immune response and inflammation. The IL-6 protein has a molecular weight of approximately 20-26 kDa. Expression of IL-6 occurs in various cell types including T cells macrophages and fibroblasts. Researchers often measure IL-6 levels in biological samples using IL-6 ELISA kits an essential tool for studying this protein's function and presence in experimental and clinical settings.
Biological function summary	IL-6 influences immune regulation and acts as part of the acute phase response. It stimulates the production of acute-phase proteins and supports the differentiation of B cells into antibody-producing cells. IL-6 is not known to be part of a larger complex acting primarily as a single entity in signal transduction. Moreover IL-6 impacts the metabolism of iron and bone homeostasis showing its multifunctional nature.
Pathways	IL-6 forms an integral part of several signaling routes particularly the JAK-STAT pathway. In this context IL-6 interacts with signal transducer proteins like STAT3 to transmit signals from the cell surface to the nucleus affecting gene expression. Another important pathway is the MAPK pathway through which IL-6 influences cell proliferation and survival. These interactions reflect IL-6's diverse effects in cellular processes.
Associated diseases and disorders	IL-6's association with rheumatoid arthritis and multiple myeloma emphasizes its role in chronic inflammation and cancer. In rheumatoid arthritis IL-6 contributes to inflammation and joint damage often together with TNF-alpha highlighting a potential target for anti-inflammatory therapies. In multiple myeloma IL-6 supports the survival and proliferation of cancerous plasma cells highlighting its importance in cancer progression and possible treatment targets.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

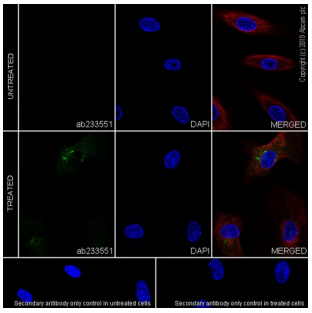
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

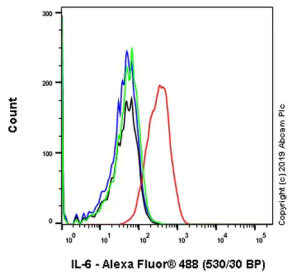
7 product images



Immunocytochemistry/ Immunofluorescence - Anti-IL-6 antibody [EPR22565-204] (ab233551)

IL-6 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-IL-6 antibody

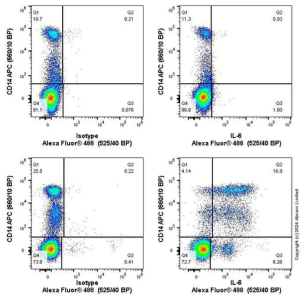
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HUVEC (human umbilical vein endothelial cell) cells labeling IL-6 with ab233551 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining in HUVEC cells treated with lipopolysaccharide (0.5µg/ml) for 4 h, then together with Brefeldin A (300ng/ml) for another 20h. The nuclear counterstain is DAPI (blue). Counterstained with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at a 1/200 dilution (red). The negative control is the secondary antibody only.



Flow Cytometry (Intracellular) - Anti-IL-6 antibody [EPR22565-204] (ab233551)

IL-6 Flow Cytometry (Intracellular) staining using rabbit Anti-IL-6 antibody

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized HUVEC (Human umbilical vein endothelial cell) treated with 0.5ug/ml LPS for 4h, then together with 300ng/ml BFA for another 20h (Red) / Untreated control (Green), labeling IL-6 with ab233551 at 1/400 (red) compared with a Rabbit monoclonal IgG (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077), at 1/2000 dilution was used as the secondary antibody.



Flow Cytometry (Intracellular) - Anti-IL-6 antibody [EPR22565-204] (ab233551)

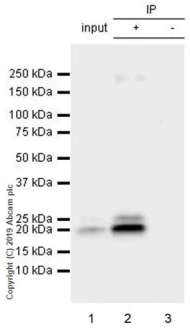
IL-6 Flow Cytometry (Intracellular) staining using rabbit Anti-IL-6 antibody

Flow cytometry staining of human peripheral blood mononuclear cells (PBMCs) (top) or PBMCs treated with LPS, 18 Hours, 1µg/mL (bottom), with ab233551 (right) or Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control (left).

PBMCs were incubated for 30 min on ice in 1x PBS containing 10 µg/ml human IgG and 10 % normal goat serum to block FC receptors and non-specific protein-protein interaction followed by incubation with the CD14 multistain. Cells were then fixed and permeabilised with BD Cytofix/Cytoperm™ for 20 min, and stained with the antibody ab233551 or Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control (1x 10^6 in 100 µl at 0.04 µg/ml (1/12)) for 30 min at 22°C.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C.

Acquisition of >30000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter. Events were gated on live cells.



Immunoprecipitation - Anti-IL-6 antibody [EPR22565-204] (ab233551)

IL-6 was immunoprecipitated from 0.35 mg HUVEC (Human umbilical vein endothelial cell) treated with 0.5µg/ml LPS for 4h, then together with 300ng/ml BFA for another 20h whole cell lysate with ab233551 at 1/20 dilution. Western blot was performed from the immunoprecipitate using ab233551 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used at 1/5000 dilution.
Lane 1: HUVEC treated as above whole cell lysate 10 µg (Input).

Lane 2: ab233551 IP in HUVEC treated as above whole cell lysate.

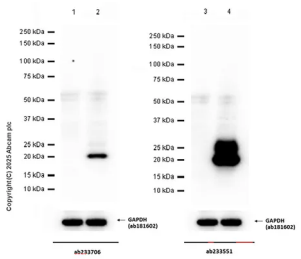
Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab233551 in HUVEC treated as above whole cell lysate.

Blocking/Dilution buffer: 5% NFDM/TBST.
Exposure time: 15 seconds.

All lanes:
Immunoprecipitation - Anti-IL-6 antibody [EPR22565-204] (ab233551)

Predicted band size: 23 kDa

Observed band size: 21 kDa, 28 kDa



Western blot - Anti-IL-6 antibody [EPR22565-204] (ab233551)

IL-6 Western blot staining using rabbit Anti-IL-6 antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST.
In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/1000000 dilution.

In Western blot analysis, ab233706 exhibited lower sensitivity compared to ab233551, for which reason we recommend the latter one as superior alternative.

ab233706 does not work in THP-1 cell line due to sensitivity issue.

Lanes 1 - 2:
Western blot - Anti-IL-6 antibody [EPR21711] (ab233706) at 1/1000 dilution

Lanes 3 - 4:
Western blot - Anti-IL-6 antibody [EPR22565-204] (ab233551) at 1/1000 dilution

Lanes 1 and 3:
Untreated HUVEC (human umbilical vein endothelial cell line) whole cell lysate at 20 µg

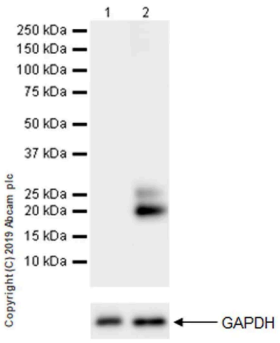
Lanes 2 and 4:
HUVEC treated with 0.5 ug/ml lipopolysaccharides (LPS) for 24 hours, added 300 ng/ml BFA last 20 hours whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Observed band size: 23 kDa

Exposure time: 20s



Western blot - Anti-IL-6 antibody [EPR22565-204] (ab233551)

The molecular weight observed is consistent with what has been described in the literature (PMID: 14970177).
Blocking/Dilution buffer: 5% NFDM/TBST.

All lanes:
Western blot - Anti-IL-6 antibody [EPR22565-204] (ab233551) at 1/1000 dilution

Lane 1:
Untreated HUVEC (human umbilical vein endothelial cell) whole cell lysate at 20 µg

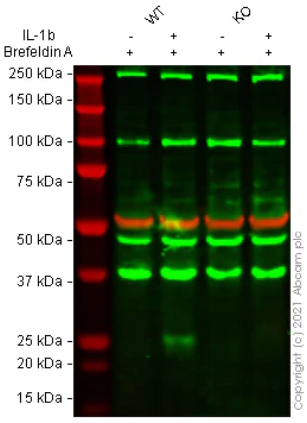
Lane 2:
HUVEC treated with 0.5 µg/ml LPS for 4 hours, then 300 ng/ml BFA (Brefeldin A) was added for the last 20 hours, whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 23 kDa

Observed band size: 21 kDa, 24 kDa



Western blot - Anti-IL-6 antibody [EPR22565-204] (ab233551)

IL-6 Western blot staining using rabbit Anti-IL-6 antibody

False colour image of Western blot: Anti-IL-6 antibody [EPR22565-204] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (ab7291) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab233551 was shown to bind specifically to IL-6. A band was observed at 25 kDa in wild-type A549 cell lysates with no signal observed at this size in IL6 knockout cell line ab273751 (knockout cell lysate ab275501). To generate this image, wild-type and IL6 knockout A549 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.
All lanes:
Western blot - Anti-IL-6 antibody [EPR22565-204] (ab233551) at 1/1000 dilution

Lane 1:
Wild-type A549 Vehicle Control IL-1b (0 ng/mL, 24 h), Brefeldin A (5 ug/mL, final 6 h) cell lysate at 30 µg

Lane 2:
Wild-type A549 Treated IL-1b (20 ng/mL, 24 h), Brefeldin A (5 ug/mL, final 6 h) cell lysate at 30 µg

Lane 3:
IL-6 knockout A549 Vehicle Control IL-1b (0 ng/mL, 24 h), Brefeldin A (5 ug/mL, final 6 h) cell lysate at 30 µg

Lane 4:
IL-6 knockout A549 Treated IL-1b (20 ng/mL, 24 h), Brefeldin A (5 ug/mL, final 6 h) cell lysate at 30 µg

Performed under reducing conditions.

Predicted band size: 23 kDa

Observed band size: 25 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.