abcam

ab283861

Anti-CD28 antibody [EPR24592-5] - BSA and Azide free

Recombinant | RabMAb

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR24592-5
Purification technique	Affinity purification Protein A
Concentration	1.018 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ICC/IF

Tested

Species	Human
Dilution info	-

Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

IP

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

WB

Not recommended

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

IHC-P

Not recommended

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

Target data

See full target information CD28

Function

Receptor that plays a role in T-cell activation, proliferation, survival and the maintenance of immune homeostasis (PubMed:1650475, PubMed:7568038). Functions not only as an amplifier of TCR signals but delivers unique signals that control intracellular biochemical events that alter the gene expression program of T-cells (PubMed:24665965). Stimulation upon engagement of its cognate ligands CD80 or CD86 increases proliferation and expression of various cytokines in particular IL2 production in both CD4(+) and CD8(+) T-cell subsets (PubMed:12196291, PubMed:1650475, PubMed:35397202). Mechanistically, ligation induces recruitment of protein kinase C-theta/PRKCQ and GRB2 leading to NF-kappa-B activation via both PI3K/Akt-dependent and -independent pathways (PubMed:21964608, PubMed:24665965, PubMed:7568038). In conjunction with TCR/CD3 ligation and CD40L costimulation, enhances the production of IL4 and IL10 in T-cells (PubMed:8617933). Isoform 3. Enhances CD40L-mediated activation of NF-kappa-B and kinases MAPK8 and PAK2 in T-cells (PubMed:15067037).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C

Notes

ab283861 is the carrier-free version of ab283860.

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.

Conjugation ready

Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Supplementary info

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

Activity summary	CD28 also known as T-cell-specific surface glycoprotein CD28 or TP44 plays an important role as a co-stimulatory receptor on T cells. This glycoprotein has a molecular mass of about 44 kDa and is located mainly on the cell surface of T cells. CD28 expression also occurs on thymocytes and some B cells. It forms part of the immunoglobulin superfamily featuring in significant antibody interactions. Researchers often use anti-CD28 antibodies for various immunological assays including CD28 ELISA to study its function.
Biological function summary	CD28 co-stimulates T cells after antigen presentation. It enhances T cell proliferation survival and cytokine production. CD28 functions alongside the T cell receptor (TCR) complex mediating signals that promote full T cell activation. This interaction amplifies the immune response forming an important axis of immune system communication. CD28 interacts with its ligands B7-1 (CD80) and B7-2 (CD86) found on antigen-presenting cells (APCs) illustrating its role in immune system dynamics.
Pathways	CD28 is integral to the PI3K/Akt and MAPK/ERK signaling pathways. These pathways conduct signals critical for cellular growth and survival linking CD28 to

	downstream transcription factors. The interaction with another protein CTLA-4 acts as a regulatory balance for CD28 signaling preventing over-activation of T cells. This balance influences immune homeostasis and tolerance providing a framework for understanding costimulatory and inhibitory signaling.
Associated diseases and disorders	CD28's role becomes evident in autoimmune diseases and transplant rejection. Abnormal CD28 signaling links to rheumatoid arthritis where inappropriate T cell activation occurs. Similarly CD28 activity contributes to organ transplant rejection by stimulating T cell-mediated responses against transplanted tissues. CTLA-4 through regulatory feedback can modify these effects providing therapeutic target strategies for managing such disorders. Efforts to modulate CD28 activity involve developing therapeutic agents such as anti-CD28 monoclonal antibodies.

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

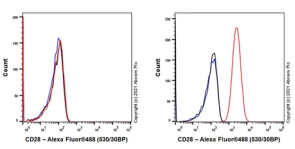
We do not recommend this combination. It is not covered by our product promise.

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.

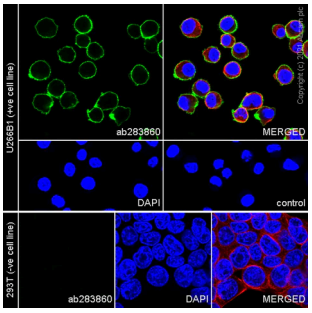
6 product images



Flow Cytometry - Anti-CD28 antibody [EPR24592-5] - BSA and Azide free (ab283861)

CD28 Flow Cytometry staining using rabbit Anti-CD28 antibody

This data was developed using ab283860, the same antibody clone in a different buffer formulation.
Flow cytometric analysis of THP-1 (Human monocytic leukemia monocyte, left) / U266B1 (human multiple myeloma B lymphocyte, right) cells labelling CD28 with ab283860 at 1/500 dilution (0.1ug)/ red compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody. Negative control: THP-1(PMID: 26773151)

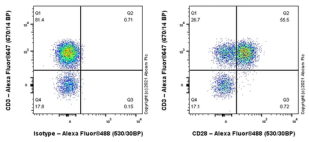


Immunocytochemistry/ Immunofluorescence - Anti-CD28 antibody [EPR24592-5] - BSA and Azide free (ab283861)

CD28 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CD28 antibody

This data was developed using ab283860, the same antibody clone in a different buffer formulation.
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized U266B1 cells labelling CD28 with ab283860 at 1/200 (2.675 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2ug/ml dilution (Green). Confocal image showing membranous staining in U266B1 cells and no staining in 293T cells. Negative control: 293T(PMID: 27708164) is observed. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 2.5ug/ml dilution (Red). The Nuclear counterstain was DAPI (Blue).

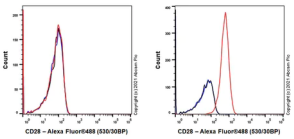
Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2ug/ml dilution.



Flow Cytometry - Anti-CD28 antibody [EPR24592-5] - BSA and Azide free (ab283861)

CD28 Flow Cytometry staining using rabbit Anti-CD28 antibody

This data was developed using ab283860, the same antibody clone in a different buffer formulation.
Flow cytometric analysis of Human peripheral blood mononuclear cell (PBMC) cells labelling CD28 with ab283860 at 1/50 dilution (1ug)/ Right compared with a Rabbit monoclonal IgG (ab172730) / Left isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody. Cells were stained with rabbit IgG or ab283860. Then stained with anti-CD3 conjugated to Alexa Fluor® 647.Gated on viable cells.

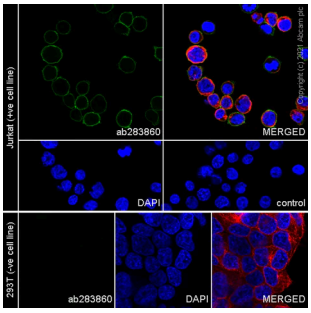


Flow Cytometry - Anti-CD28 antibody [EPR24592-5] - BSA and Azide free (ab283861)

CD28 Flow Cytometry staining using rabbit Anti-CD28 antibody

This data was developed using ab283860, the same antibody clone in a different buffer formulation.
Flow cytometric analysis of 293T (Human embryonic kidney epithelial cell, left) / Jurkat (Human T cell leukemia T lymphocyte, right) cells labelling CD28 with ab283860 at 1/500 dilution (0.1ug)/ red compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). Goat

Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody. Negative control: 293T(PMID: 27708164)

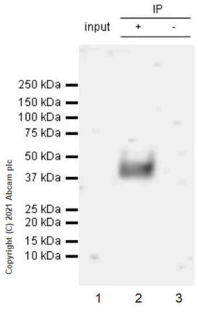


Immunocytochemistry/ Immunofluorescence - Anti-CD28 antibody [EPR24592-5] - BSA and Azide free (ab283861)

CD28 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CD28 antibody

This data was developed using ab283860, the same antibody clone in a different buffer formulation.
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized Jurkat cells labelling CD28 with ab283860 at 1/200 (2.675 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2ug/ml dilution (Green). Confocal image showing membranous staining in Jurkat cells and no staining in 293T cells. Negative control: 293T(PMID: 27708164) is observed. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 2.5ug/ml dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2ug/ml dilution.



Immunoprecipitation - Anti-CD28 antibody [EPR24592-5] - BSA and Azide free (ab283861)

This data was developed using ab283860, the same antibody clone in a different buffer formulation.
CD28 was immunoprecipitated from 0.35 mg Jurkat (human T cell leukemia cell line from peripheral blood) whole cell lysate 10 ug with ab283860 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab283860 at 1/1000 dilution. VeriBlot for IP secondary antibody (HRP) (ab131366) was used at 1/5000 dilution.

Lane 1: Jurkat (human T cell leukemia cell line from peripheral blood) whole cell lysate 10 ug

Lane 2: ab283860 IP in Jurkat whole cell lysate

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab283860 in Jurkat whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 3 minutes

This blot was developed using a higher sensitivity ECL substrate.

All lanes:
Immunoprecipitation - Anti-CD28 antibody [EPR24592-5] (ab283860)

Predicted band size: 25 kDa

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