abcam

ab133600

Anti-CBFb antibody [EPR6322]

Recombinant | RabMAb | KO Validated

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	EPR6322
Purification technique	Affinity purification Protein A
Dissociation constant	7.2×10^{-11} M
Concentration	1.25 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Not recommended

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

WB

Tested

Species	Mouse
Dilution info	1/1000
Notes	-

Species	Rat
Dilution info	1/1000
Notes	-

Species	Human
Dilution info	1/1000
Notes	-

IHC-P

Not recommended

Species	Mouse
Dilution info	-
Notes	**For unpurified use at 1/100 - 1/250.** Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Rat
Dilution info	-
Notes	**For unpurified use at 1/100 - 1/250.** Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Human
Dilution info	-
Notes	**For unpurified use at 1/100 - 1/250.** Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

ICC/IF

Tested

Species	Human
Dilution info	1/100 - 1/600
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/100 - 1/10000
Notes	For unpurified use at 1/190. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information CBFB

Function	Forms the heterodimeric complex core-binding factor (CBF) with RUNX family proteins (RUNX1, RUNX2, and RUNX3). RUNX members modulate the transcription of their target genes through recognizing the core consensus binding sequence 5'-TGTGGT-3', or very rarely, 5'-TGCGGT-3', within their regulatory regions via their runt domain, while CBFB is a non-DNA-binding regulatory subunit that allosterically enhances the sequence-specific DNA-binding capacity of RUNX. The heterodimers bind to the core site of a number of enhancers and promoters, including murine leukemia virus, polyomavirus enhancer, T-cell receptor

enhancers, LCK, IL3 and GM-CSF promoters. CBF complexes repress ZBTB7B transcription factor during cytotoxic (CD8+) T cell development. They bind to RUNX-binding sequence within the ZBTB7B locus acting as transcriptional silencer and allowing for cytotoxic T cell differentiation. (Microbial infection) Following infection, hijacked by the HIV-1 Vif protein, leading to the formation a cullin-5-RING E3 ubiquitin-protein ligase complex (ECS complex) that catalyzes ubiquitination and degradation of APOBEC3F and APOBEC3G (PubMed:22190037, PubMed:31792451, PubMed:36598981, PubMed:36754086, PubMed:37419875). The complex can also ubiquitinate APOBEC3H to some extent (PubMed:37640699). Association with HIV-1 Vif protein also inhibits the transcription coactivator activity of CBFB/CBF-beta (PubMed:22190037).

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	CBFb also known as Core-Binding Factor Subunit Beta weighs approximately 22 kDa. It plays an important role in gene regulation by stabilizing the DNA binding of core-binding factor alpha (CBFA) subunits such as RUNX proteins. CBFb does not bind DNA directly. Instead it enhances the affinity of the RUNX family for their DNA targets. CBFb is broadly expressed in various tissues with high levels observed in hematopoietic cells.
Biological function summary	CBFb forms a complex with RUNX transcription factors facilitating their function. The complex is essential for hematopoiesis as it regulates the expression of genes critical for blood cell differentiation and proliferation. CBFb is involved in bone development influencing the formation of bone-related genes. This CBFb/RUNX complex plays a role in regulating the cell cycle and apoptosis fundamental processes for maintaining cellular integrity.
Pathways	More than one signaling pathway includes CBFb's involvement. Particularly it participates in the Wnt signaling pathway an important player in cell differentiation. Additionally CBFb/RUNX complex regulates genes downstream in the TGF-beta signaling pathway impacting cellular proliferation and differentiation. Through these pathways CBFb is linked to proteins like SMAD3 further emphasizing its role in mediating various biological processes.
Associated diseases and disorders	Abnormalities in CBFb function connect to acute myeloid leukemia (AML). CBFb's interaction with RUNX1 can result in a chromosomal translocation that generates the CBFb-MYH11 fusion protein which disrupts normal hematopoiesis and contributes to leukemogenesis. Moreover CBFb variations associate with cleidocranial dysplasia a skeletal disorder through its interaction with the RUNX2 protein highlighting its significance in bone development.

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

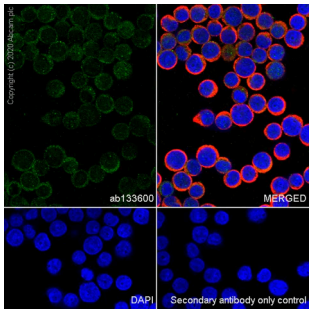
We do not recommend this combination. It is not covered by our product promise.

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.

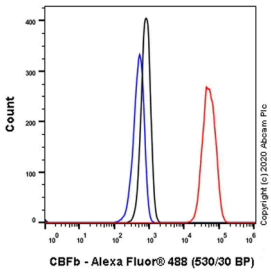
6 product images



Immunocytochemistry/ Immunofluorescence - Anti-CBFb antibody [EPR6322] (ab133600)

CBFb Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CBFb antibody

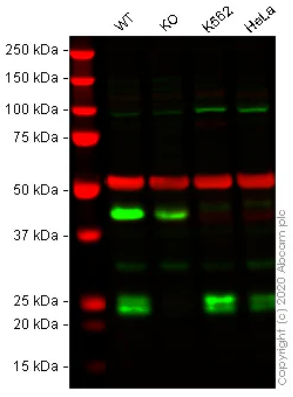
Immunocytochemistry/Immunofluorescence analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labeling CBFb with purified ab133600 at 1/100 dilution (10 µg/mL). Cells were fixed in 100% Methanol and permeabilized with None. Cells were counterstained with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5 µg/mL). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1/1000 (2 µg/mL) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Flow Cytometry (Intracellular) - Anti-CBFb antibody [EPR6322] (ab133600)

CBFb Flow Cytometry (Intracellular) staining using rabbit Anti-CBFb antibody

Intracellular Flow Cytometry analysis of K-562 (Human chronic myelogenous leukemia lymphoblast) cells, labeling CBFb with purified ab133600 at 1/100 dilution (10µg/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor®488, ab150077) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Western blot - Anti-CBFb antibody [EPR6322] (ab133600)

CBFb Western blot staining using rabbit Anti-CBFb antibody

Lanes 1 - 4: Merged signal (red and green). Green - ab133600 observed at 22 kDa. Red - loading control ab7291 (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.
ab133600 was shown to react with CBFb in wild-type A431 cells in western blot with loss of signal observed in CBFB knockout sample. Wild-type and CBFB knockout A431 cell lysates were subjected to SDS-PAGE. Membranes were blocked in 2% BSA in TBS-T (0.1% Tween®) before incubation with ab133600 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.

All lanes:
Western blot - Anti-CBFb antibody [EPR6322] (ab133600) at 1/1000 dilution

Lane 1:
Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate at 20 µg

Lane 2:
Western blot - Human CBFB knockout A-431 cell lysate (ab270495) at 20 µg

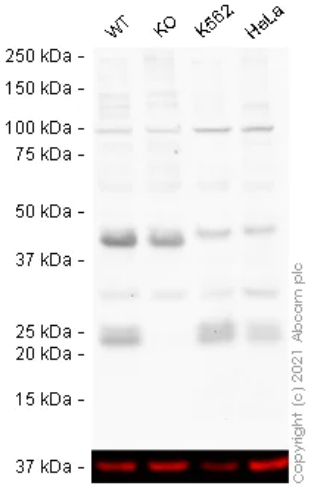
Lane 3:
K562 (Human chronic myelogenous leukemia lymphoblast cell line) whole cell lysate at 20 µg

Lane 4:
HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 22 kDa

Observed band size: 22 kDa



Western blot - Anti-CBFb antibody [EPR6322] (ab133600)

CBFb Western blot staining using rabbit Anti-CBFb antibody

ab133600 was shown to react with CBFb in wild-type A431 cells in western blot. Loss of signal was observed when CBFB knockout sample was used. Membranes were blocked in 2 % BSA in TBS-T (0.1 % Tween®) before incubation with ab133600 overnight at 4°C at a 1 in 1000 dilution and ab184095 (Mouse Anti-GAPDH antibody [mAbcam 9484] - Alexa Fluor® 680) at a 1 in 1000 dilution. Blots were incubated with HRP conjugated Goat anti-Rabbit (H+L) secondary antibody at 1/5000 for 1 hour at room temperature before development with Optiblot ECL reagent (ab133456) and imaging.
All lanes:
Western blot - Anti-CBFb antibody [EPR6322] (ab133600) at 1/1000 dilution

Lane 1:
Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate at 20 µg

Lane 2:
CBFB knockout A-431 (Human epidermoid carcinoma cell line) whole cell lysate at 20 µg

Lane 2:
Western blot - Human CBFB knockout A-431 cell line (ab270472)

Lane 3:
K562 (Human chronic myelogenous leukemia lymphoblast cell line) whole cell lysate at 20 µg

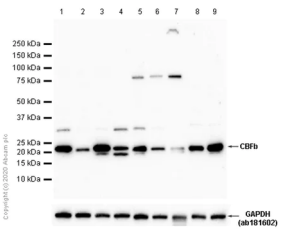
Lane 4:
HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 22 kDa

Observed band size: 24 kDa

Exposure time: 150s



Western blot - Anti-CBFb antibody [EPR6322] (ab133600)

Blocking/Diluting buffer: 5% NFDM/TBST
Loading Control: Rabbit monoclonal [EPR16891] to GAPDH (ab181602)

All lanes:
Western blot - Anti-CBFb antibody [EPR6322] (ab133600) at 1/1000 dilution

Lane 1:
Raji (Human Burkitt's lymphoma B lymphocyte) whole cell lysate at 20 µg

Lane 2:
HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 3:
K-562 (Human chronic myelogenous leukemia lymphoblast) whole cell lysate at 20 µg

Lane 4:
Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate at 20 µg

Lane 5:
RAW 264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate at 20 µg

Lane 6:
Mouse spleen lysate at 20 µg

Lane 7:
Mouse thymus lysate at 20 µg

Lane 8:
Rat spleen lysate at 20 µg

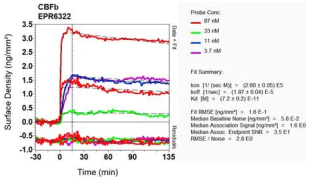
Lane 9:
Rat thymus lysate at 20 µg

Secondary

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

Predicted band size: 22 kDa

Observed band size: 22 kDa



OI-RD Scanning - Anti-CBFb antibody [EPR6322] (ab133600)

We have systematically measured KD (the equilibrium dissociation constant between the antibody and its antigen), of more than 840 recombinant antibodies to assess not only their individual KD values but also to see the average affinity of antibody. Based on the comparison with published literature values for mouse monoclonal antibodies, Recombinant antibodies appear to be on average 1-2 order of magnitude higher affinity.

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