abcam

Product datasheet

# Anti-CBFb antibody [EPR6322] ab133600

KO VALIDATED Recombinant RabMAb

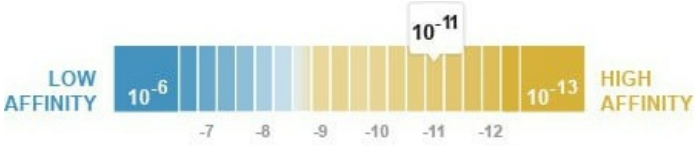
5 References 7 Images

## Overview

| | |
|---|---|
| **Product name** | Anti-CBFb antibody [EPR6322] |
| **Description** | Rabbit monoclonal [EPR6322] to CBFb |
| **Host species** | Rabbit |
| **Tested applications** | **Suitable for:** WB, ICC/IF, Flow Cyt (Intra)<br>**Unsuitable for:** IHC-P or IP |
| **Species reactivity** | **Reacts with:** Mouse, Rat, Human |
| **Immunogen** | Synthetic peptide within Human CBFb aa 100-200. The exact sequence is proprietary. |
| **Positive control** | WB: A431, Raji, HeLa, K562, Jurkat, and Raw264.7 cell lysates. Mouse and rat spleen and thymus tissue lysates; ICC/IF: Jurkat cells; Flow Cyt (intra): K562 cells. |
| **General notes** | This product is a recombinant monoclonal antibody, which offers several advantages including:<br>- High batch-to-batch consistency and reproducibility<br>- Improved sensitivity and specificity<br>- Long-term security of supply<br>- Animal-free production<br>For more information **see here**.<br>Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**. |

## Properties

| | |
|---|---|
| **Form** | Liquid |
| **Storage instructions** | 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. |
| **Dissociation constant ($K_D$)** | $K_D = 7.20 \times 10^{-11}$ M |




Learn more about $K_D$

| | |
|---|---|
| **Storage buffer** | pH: 7.2 |

| | |
|---|---|
| | Preservative: 0.01% Sodium azide<br>Constituents: 0.05% BSA, 59% PBS, 40% Glycerol |
| **Purity** | Protein A purified |
| **Clonality** | Monoclonal |
| **Clone number** | EPR6322 |
| **Isotype** | IgG |

## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab133600 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

| Application | Abreviews | Notes |
|---|---|---|
| **WB** | | 1/1000. Predicted molecular weight: 22 kDa. |
| **ICC/IF** | | 1/100 - 1/600. |
| **Flow Cyt (Intra)** | | 1/100 - 1/10000.<br>For unpurified use at 1/190. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. |

**Application notes** Is unsuitable for IHC-P or IP.

## Target

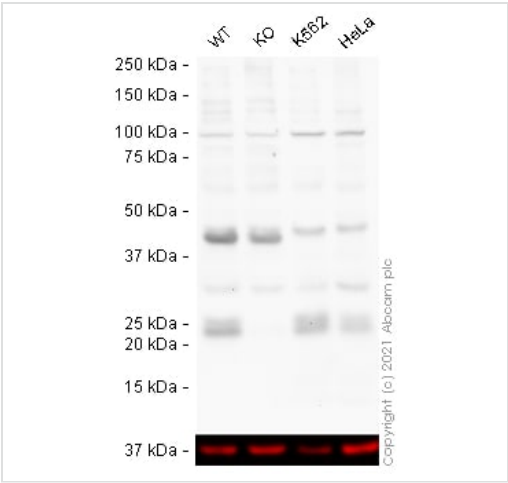
**Function** CBF binds to the core site, 5'-PYGPYGGT-3', of a number of enhancers and promoters, including murine leukemia virus, polyomavirus enhancer, T-cell receptor enhancers, LCK, IL3 and GM-CSF promoters. CBFB enhances DNA binding by RUNX1.

**Involvement in disease** Note=A chromosomal aberration involving CBFB is associated with acute myeloid leukemia of M4EO subtype. Pericentric inversion inv(16)(p13;q22). The inversion produces a fusion protein that consists of the 165 N-terminal residues of CBF-beta (PEPB2) with the tail region of MYH11.

**Sequence similarities** Belongs to the CBF-beta family.

**Cellular localization** Nucleus.

## Images




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

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

**Lane 1 :** Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate
**Lane 2 :** CBFB knockout A-431 (Human epidermoid carcinoma cell line) whole cell lysate
**Lane 3 :** K562 (Human chronic myelogenous leukemia lymphoblast cell line ) whole cell lysate
**Lane 4 :** HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

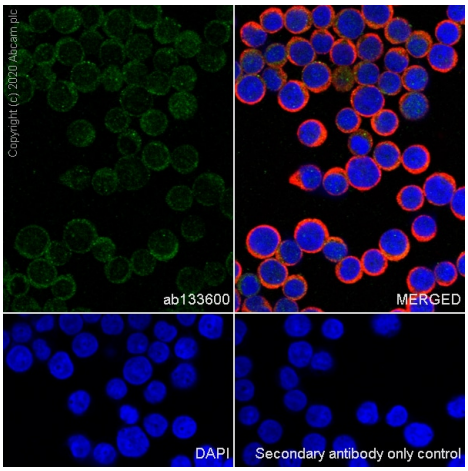
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 22 kDa
**Observed band size:** 24 kDa

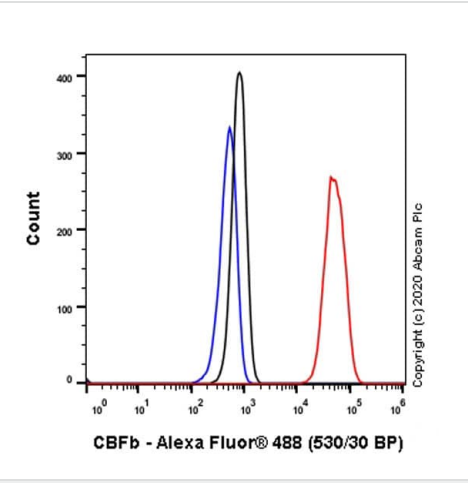
**Exposure time:** 150 seconds

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.



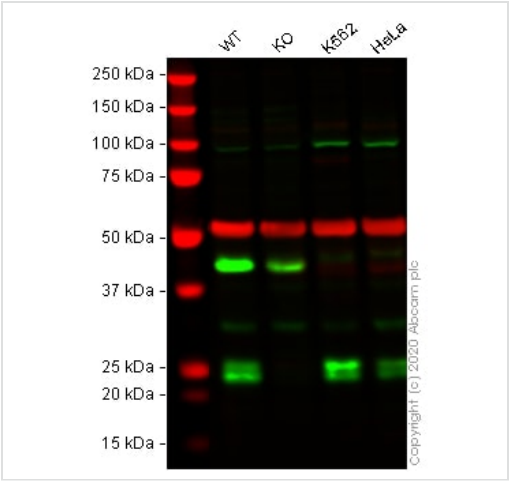

Immunocytochemistry/ Immunofluorescence - Anti-CBFb antibody [EPR6322] (ab133600)

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)

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)

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

**Lane 1 :** Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate
**Lane 2 :** CBFB knockout A-431 (Human epidermoid carcinoma cell line) whole cell lysate
**Lane 3 :** K562 (Human chronic myelogenous leukemia lymphoblast cell line ) whole cell lysate
**Lane 4 :** HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

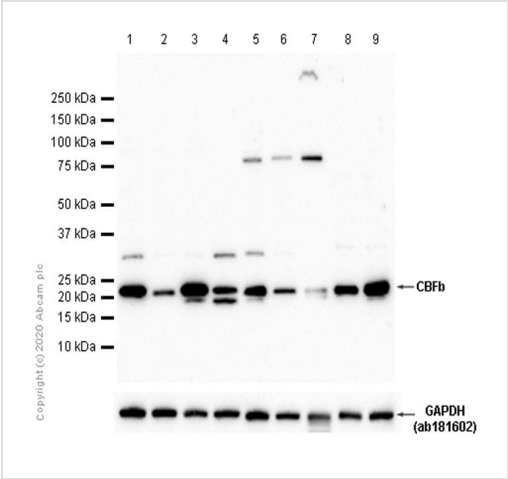
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 22 kDa
**Observed band size:** 22 kDa

**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.




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

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

**Lane 1 :** Raji (Human Burkitt's lymphoma B lymphocyte) whole cell lysate
**Lane 2 :** HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate
**Lane 3 :** K-562 (Human chronic myelogenous leukemia lymphoblast) whole cell lysate
**Lane 4 :** Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate
**Lane 5 :** RAW 264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate
**Lane 6 :** Mouse spleen lysate
**Lane 7 :** Mouse thymus lysate
**Lane 8 :** Rat spleen lysate
**Lane 9 :** Rat thymus lysate

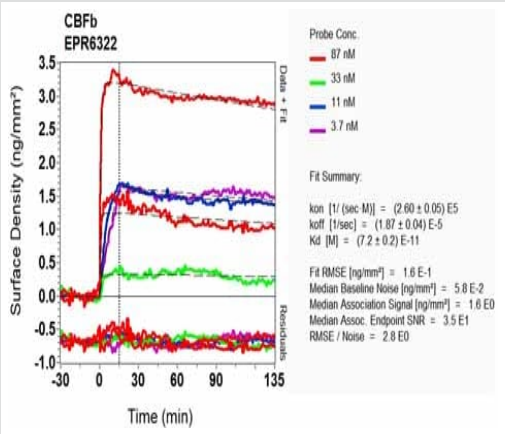
Lysates/proteins at 20 µg per lane.

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

**Predicted band size:** 22 kDa
**Observed band size:** 22 kDa

**Blocking/Diluting buffer:** 5% NFDM/TBST

**Loading Control:** Rabbit monoclonal [EPR16891] to GAPDH (**ab181602**)




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

Equilibrium disassociation constant ($K_D$)

Learn more about $K_D$

**Click here to learn more about $K_D$**


Anti-CBFb antibody [EPR6322] (ab133600)



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