abcam

ab32027

Anti-CBL antibody [YE323] - C-terminal

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	YE323
Purification technique	Affinity purification Protein A
Epitope	ab32027 reacts with an epitope located in the C terminal region of CBL.
Concentration	0.213 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ICC/IF

Tested

Species	Human
Dilution info	1/50
Notes	**For unpurified use at 1/100**

Expected

Species	Mouse
Dilution info	1/50
Notes	**For unpurified use at 1/100**

Species	Rat
Dilution info	1/50
Notes	**For unpurified use at 1/100**

Predicted

Species	Chicken
Dilution info	-
Notes	-

IP

Not recommended

Species	Mouse, Rat, Human, Chicken
Dilution info	-
Notes	-

WB

Tested

Species	Mouse
Dilution info	1/1000
Notes	**For unpurified use at 1/5000**

Species	Human
Dilution info	1/1000
Notes	**For unpurified use at 1/5000**

Species	Rat
Dilution info	1/1000
Notes	**For unpurified use at 1/5000**

Predicted

Species	Chicken
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/30
Notes	For unpurified use at 1/100 ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse
Dilution info	1/30
Notes	For unpurified use at 1/100 ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Species	Rat
Dilution info	1/30
Notes	For unpurified use at 1/100 ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Predicted

Species	Chicken
Dilution info	-
Notes	-

Target data

See full target information CBL

Function	E3 ubiquitin-protein ligase that acts as a negative regulator of many signaling pathways by mediating ubiquitination of cell surface receptors (PubMed:10514377, PubMed:11896602, PubMed:14661060, PubMed:14739300, PubMed:15190072, PubMed:17509076, PubMed:18374639, PubMed:19689429, PubMed:21596750, PubMed:28381567, PubMed:40101708). Accepts ubiquitin from specific E2 ubiquitin-conjugating enzymes, and then transfers it to substrates promoting their degradation by the proteasome (PubMed:10514377, PubMed:14661060, PubMed:14739300, PubMed:17094949, PubMed:17509076, PubMed:17974561). Recognizes activated receptor tyrosine kinases, including KIT, FLT1, FGFR1, FGFR2, PDGFRA, PDGFRB, CSF1R, EPHA8 and KDR and mediates their ubiquitination to terminate signaling (PubMed:15190072, PubMed:18374639, PubMed:21596750). Recognizes membrane-bound HCK, SRC and other kinases of the SRC family and mediates their ubiquitination and degradation (PubMed:11896602). Ubiquitinates EGFR and SPRY2 (PubMed:17094949, PubMed:17974561). Involved in LAG3-mediated inhibition of TCR signaling: following ligand-binding to LAG3, catalyzes 'Lys-63'-linked ubiquitination of LAG3, unleashing the LAG3 C-terminus from the membrane, and initiating a signaling that prevents TCR activation (PubMed:40101708). Ubiquitinates NECTIN1 following association between NECTIN1 and herpes simplex virus 1/HHV-1 envelope glycoprotein D, leading to NECTIN1 removal from cell surface (PubMed:28381567). Participates in signal transduction in hematopoietic cells. Plays an important role in the regulation of osteoblast differentiation and apoptosis (PubMed:15190072, PubMed:18374639). Essential for osteoclastic bone resorption (PubMed:14739300). The 'Tyr-731' phosphorylated form induces the activation and recruitment of phosphatidylinositol 3-kinase to the cell membrane in a signaling pathway that is critical for osteoclast function (PubMed:14739300). In association with CBLB, required for proper feedback inhibition of ciliary platelet-derived growth factor receptor-alpha (PDGFRA) signaling pathway via ubiquitination and internalization of PDGFRA (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	The CBL protein also known as C-cbl or Casitas B-lineage lymphoma proto-oncogene serves as an E3 ubiquitin-protein ligase. This protein with a molecular mass of approximately 120-130 kDa plays a critical role in the regulation of receptor tyrosine kinases. It facilitates the polyubiquitination of activated receptor proteins targeting them for degradation in the proteasome. CBL is predominantly expressed in various tissues including the hematopoietic system. It is a significant component in the control of cell signaling and helps maintain cellular homeostasis.
Biological function summary	The CBL protein acts as a negative regulator for several signaling pathways. It integrates into signaling complexes and modulates the activation of pathways by interacting with other proteins. CBL does not act alone; it commonly forms complexes with proteins such as Grb2 and p85 influencing downstream signaling influence in pathways. This regulation prevents excessive activation of signals that can lead to cellular proliferation or differentiation abnormally.
Pathways	CBL integrates into the ubiquitin-proteasome system and the Ras signaling pathway. Within these pathways CBL works to modulate cell surface receptors ensuring that signaling threshold is not surpassed. The importance of CBL in the Ras signaling pathway is highlighted by its interactions with proteins like Ras-GAP and SHP2. Through these interactions CBL controls the rate of signal transduction affecting cellular responses such as growth survival and motility.
Associated diseases and disorders	CBL involvement has been linked to several forms of cancer and immune deficiencies. Mutations or dysregulation of the CBL gene can lead to myelodysplastic syndromes and acute myeloid leukemia. In these conditions the interaction between CBL and other proteins such as JAK2 becomes dysregulated driving abnormal cell growth and survival. Furthermore CBL's failure to properly degrade tyrosine kinase receptors can contribute to enhanced signaling fostering oncogenic processes.

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

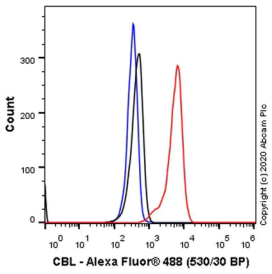
We do not recommend this combination. It is not covered by our product promise.

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.

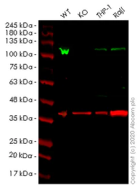
6 product images



Flow Cytometry (Intracellular) - Anti-CBL antibody [YE323] - C-terminal (ab32027)

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

Intracellular Flow Cytometry analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labeling CBL with purified ab32027 at 1/30 dilution (10 µg/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-CBL antibody [YE323] - C-terminal (ab32027)

CBL Western blot staining using rabbit Anti-CBL antibody

Lanes 1-4: Merged signal (red and green). Green - ab32027 observed at 110 kDa. Red - loading control ab8245 observed at 36 kDa.
Unpurified ab32027 Anti-CBL antibody [YE323] - C-terminal was shown to specifically react with CBL in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab267245

(knockout cell lysate ab257200) was used. Wild-type and CBL knockout samples were subjected to SDS-PAGE. ab32027 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.
All lanes:
Western blot - Anti-CBL antibody [YE323] - C-terminal (ab32027) at 1/1000 dilution

Lane 1:
Wild-type HEK293T cell lysate at 20 µg

Lane 2:
CBL knockout HEK293T cell lysate at 20 µg

Lane 3:
THP-1 cell lysate at 20 µg

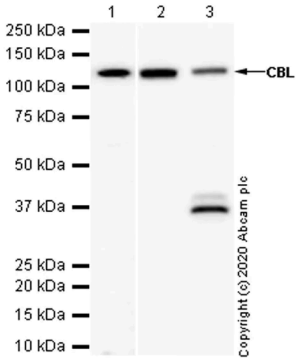
Lane 4:
Raji cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Predicted band size: 100 kDa

Observed band size: 110 kDa



Western blot - Anti-CBL antibody [YE323] - C-terminal (ab32027)

CBL Western blot staining using rabbit Anti-CBL antibody

All lanes:
Western blot - Anti-CBL antibody [YE323] - C-terminal (ab32027) at 1/1000 dilution

Lane 1:
Raji (Human Burkitt's lymphoma B lymphocyte) whole cell lysate at 15 µg

Lane 2:
Rat testis lysate at 20 µg

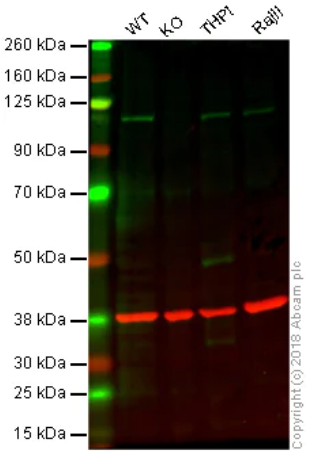
Lane 3:
Rat thymus lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/2000 dilution

Predicted band size: 100 kDa

Observed band size: 110 kDa



Western blot - Anti-CBL antibody [YE323] - C-terminal (ab32027)

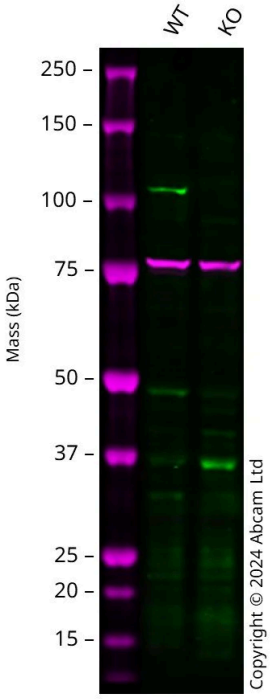
CBL Western blot staining using rabbit Anti-CBL antibody

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)
Lane 2: CBL knockout HAP1 whole cell lysate (20 µg)
Lane 3: THP1 whole cell lysate (20 µg)
Lane 4: Raji whole cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab32027 observed at 100 kDa. Red - loading control, ab9484, observed at 37 kDa.

Unpurified ab32027 was shown to specifically react with CBL in wild-type HAP1 cells as signal was lost in CBL knockout cells. Wild-type and CBL knockout samples were subjected to SDS-PAGE. ab32027 and ab9484 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/5000 dilution and 1/20000 dilution respectively. Blots were developed 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 1/20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-CBL antibody [YE323] - C-terminal (ab32027)

Predicted band size: 100 kDa



Western blot - Anti-CBL antibody [YE323] - C-terminal (ab32027)

Western blot: Anti-CBL antibody [YE323] (ab32027) staining at 1/1000 dilution, shown in green; Mouse anti-CANX [CANX/1543] (ab238078) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab32027 was shown to bind specifically to CBL. A band was observed at 105 kDa in wild-type HCT 116 cell lysates with no signal observed at this size in CBL knockout cell line. To generate this image, wild-type and CBL knockout HCT 116 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) 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 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.
All lanes:
Western blot - Anti-CBL antibody [YE323] - C-terminal (ab32027) at 1/1000 dilution

Lane 1:
Wild-type HCT 116 cell lysate at 20 µg

Lane 2:
CBL knockout HCT 116 cell lysate at 20 µg

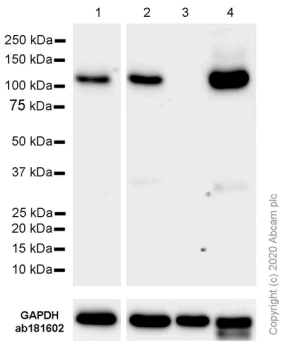
Lane 2:
Western blot - Human CBL knockout HCT116 cell line (ab287255)

Secondary

All lanes:
Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

Performed under reducing conditions.

Observed band size: 105 kDa



Western blot - Anti-CBL antibody [YE323] - C-terminal (ab32027)

Blocking and diluting buffer and concentration: 5% NFDM/TBST
Exposure time: 30 seconds

All lanes:
Western blot - Anti-CBL antibody [YE323] - C-terminal (ab32027) at 1/1000 dilution

Lane 1:
WEHI-231 (Mouse B cell lymphoma B lymphocyte) cell lysate at 10 µg

Lane 2:
F9 (Mouse embryonal carcinoma epithelial cell) cell lysate at 10 µg

Lane 3:
NIH/3T3 (Mouse embryonic fibroblast) cell lysate at 10 µg

Lane 4:
Mouse thymus lysate at 10 µg

Secondary

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

Predicted band size: 100 kDa

Observed band size: 110 kDa

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