

Technical Data Sheet

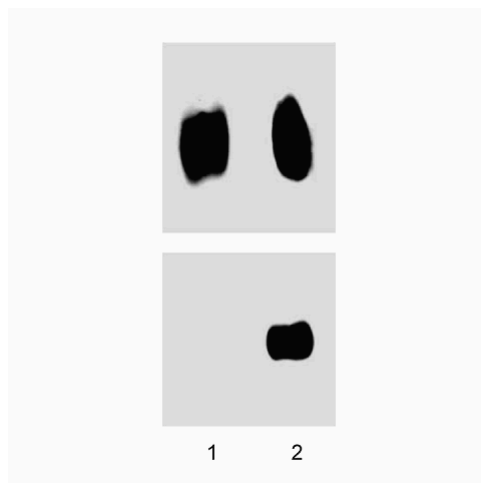
Purified Anti-c-Cbl (pY700)**Product Information**

Material Number:	612305
Size:	150 µg
Concentration:	250 µg/ml
Clone:	47/c-Cbl
Immunogen:	Human c-Cbl (pY700) Peptide
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Human
Target MW:	120 kDa
Storage Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09% sodium azide.

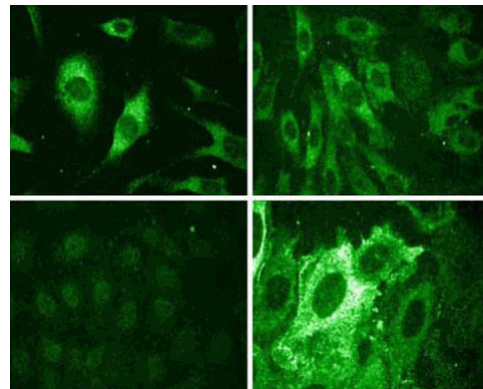
Description

Cbl (Casitas *B*-lineage lymphoma) was identified in the genome of a transforming retrovirus from a mouse pre-B lymphoma. The cellular gene product c-Cbl is one of numerous Cbl-related proteins found in vertebrate and invertebrate organisms. It is an 120-kDa adapter protein that contains multiple functional domains, including a RING finger motif, a tyrosine kinase-binding (TKB) domain, and a proline-rich region. The TKB domain directly interacts with specific auto-phosphorylation sites in activated protein-tyrosine kinases (PTK). Through the RING finger motif, c-Cbl recruits and activates an E2 ubiquitin-conjugating enzyme, thus targeting the activated PTK for protein degradation. The proline-rich region contains SH3 domain-binding and 14-3-3 protein-binding motifs. c-Cbl is also phosphorylated at tyrosines 700 (Y700), 731, and 774 by Syk- and Src-family kinases after the stimulation of some integrins and a wide variety of receptors for antigens, immunoglobulins, growth factors, cytokines, and hormones. In turn, the phosphorylated Y700 site interacts with the SH2 domains of CRK and Vav1. The c-Cbl adapter protein is expressed in the cytoplasm in all tissues, with especially high levels of expression in hematopoietic cells. Through its many functional sites, c-Cbl plays key roles in the positive and negative regulation of vital cell functions, including T Cell Receptor-mediated cellular immune responses.

The 47/c-Cbl monoclonal antibody recognizes the Y700-phosphorylated form of human c-Cbl. The orthologous phosphorylation site in mouse c-Cbl is Y698.



Jurkat cells were treated with 1mM pervanadate for 15 minutes at 37°C. The top panel was probed with c-Cbl (Cat. No. 610441) and the bottom panel was probed with c-Cbl (pY700) (Cat. No. 612304).



Eahy cells were serum starved and treated with pervanadate (1mM) for 15 minutes, then fixed in 3.75% paraformaldehyde with 0.2% Triton X-100. Immunofluorescent staining was performed using c-Cbl (Cat. No. 610441) and c-Cbl (pY700) (Cat. No. 612304).

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Store undiluted at -20°C.

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Application Notes

Application

Western blot	Routinely Tested
Immunofluorescence	Tested During Development

Recommended Assay Procedure:

Western blot: Please refer to http://www.bdbiosciences.com/pharmingen/protocols/Western_Blotting.shtml.

Suggested Companion Products

Catalog Number	Name	Size	Clone
611451	Jurkat Cell Lysate	500 µg	(none)
611755	Jurkat + Pervanadate Lysate	500 µg	(none)
610441	Purified Mouse Anti-c-Cbl	50 µg	17/c-Cbl

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

References

Donovan JA, Wange RL, Langdon WY, Samelson LE. The protein product of the c-cbl protooncogene is the 120-kDa tyrosine-phosphorylated protein in Jurkat cells activated via the T cell antigen receptor. *J Biol Chem.* 1994; 269(37):22921-22924.(Biology)
Rivero-Lezcano OM, Sameshima JH, Marcilla A, Robbins KC. Physical association between Src homology 3 elements and the protein product of the c-cbl proto-oncogene. *J Biol Chem.* 1994; 269(26):17363-17366.(Biology)