Product Information

Anti-phospho-c-Abl [pTyr^{412}]

produced in rabbit, affinity isolated antibody

C5240

Product Description

Anti-phospho-c-Abl [pTyr^{412}] is produced in rabbit using as immunogen a synthetic phosphorylated peptide derived from the region of c-Abl that contains tyrosine 412. There are two widely expressed forms of c-Abl produced by alternative splicing, known as 1a and 1b. The tyrosine 412 is the phosphorylation site of form 1b (the more commonly used form). The corresponding phosphorylation site in 1a is tyrosine 393. The antiserum is affinity purified using epitope specific affinity chromatography. The antibody is preadsorbed to remove any reactivity toward a non-phosphorylated peptide.

Anti-phospho-c-Abl [pTyr^{412}] specifically recognizes c-Abl phosphorylated at tyrosine 412 (~140-150 kDa). The antibody detects human c-Abl. Mouse c-Abl (100% homology) is expected to cross react. The antibody has been used in immunoblotting applications.

c-Abl is a 140-150 kDa non-receptor protein tyrosine kinase whose precise functions are not known, but it appears to play a role in growth factor and integrin signaling, cell cycle regulation, cytoskeletal reorganization, neurogenesis, and in responses to DNA damage and oxidative stress. c-Abl kinase activity is increased in vivo by diverse physiological stimuli including ionizing radiation, entry into S phase, integrin activation, and platelet-derived growth factor (PDGF) stimulation. c-Abl contains various protein binding domains that appear to enable it to regulate the functions of many proteins by forming complexes, most notably three isoforms of the oncogenic protein BCR/ABL.

c-Abl becomes fully activated by sequential phosphorylation of tyrosines 412 and 245.

Reagent

Supplied as a solution in Dulbecco's phosphate buffered saline pH 7.3, 50% glycerol with 1.0 mg/mL BSA and 0.05% Sodium azide as a preservative.

Precautions and Disclaimer

This product is for R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

Store at -20 °C. For extended storage, freeze in working aliquots. Repeated freezing and thawing, or storage in "frost-free" freezers, is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilutions should be discarded if not used within 12 hours.

Product Profile

The supplied reagent is sufficient for 10 blots.

Immunoblotting: a working antibody dilution of 1:1000 is recommended using fibroblasts transfected with oncogenic δSH3-Abl.

Note: In order to obtain the best results using various techniques and preparations, we recommend determining the optimal working dilutions by titration.

Results

Peptide Competition

1. Fibroblasts transfected with δSH3-Abl were resolved by SDS-PAGE on a 10% polyacrylamide gel and transferred to PVDF.
2. Membranes were blocked with a 5% BSA-TBST buffer overnight at 4 °C.
3. After blocking, membranes were preincubated with different peptides as follows:
   - Lane 1: No peptide
   - Lane 2: Non-phosphorylated peptide corresponding to the immunogen
   - Lane 3: A generic phosphotyrosine containing peptide
   - Lane 4: Immunogen
4. After preincubation membranes were incubated with 0.50 µg/mL c-Abl [pTyr^{412}] antibody for two hours at room temperature in a 3% BSA-TBST buffer.
5. After washing, membranes were incubated with goat F(ab')$_2$ anti-rabbit IgG alkaline phosphatase and signals were detected.

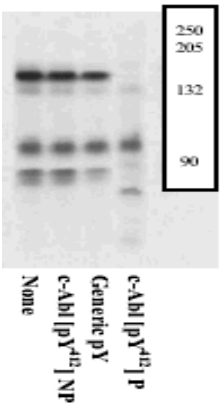

Figure 1 Peptide Competition

The data in Figure 1 show that only the peptide corresponding to c-Abl [pTyr^{412}] blocks the antibody signal, thereby demonstrating the specificity of the antibody.

References

1. Cong, F., et al. Interaction between UV-damaged DNA binding activity proteins and the c-Abl tyrosine kinase. *J. Biol. Chem*., **277**, 34870-34878 (2002).
2. Furstoss, O., et al., c-Abl is an effector of Src for growth factor-induced c-myc expression and DNA synthesis. *EMBO J*., **21**, 514-524 (2002).
3. Brasher, B.B. and R.A. Van Etten c-Abl has high intrinsic tyrosine kinase activity that is stimulated by mutation of the src homology 3 domain and by autophosphorylation at two distinct regulatory tyrosines. *J. Biol. Chem*., **275**, 35631-35637 (2000).
4. Plattner, R., et al. c-Abl is activated by growth factors and Src family kinases and has a role in the cellular response to PDGF. *Genes Dev*. **13**, 2400-2411 (1999).

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