

p-ZAP-70 (Tyr 493): sc-101823

BACKGROUND

The activation of T lymphocytes by antigens is mediated by the T cell receptor (TCR), which is a multisubunit complex assembled from at least six different genes. The TCR subunits include the α and β chains, the CD3 γ , δ and ϵ chains and a ζ -containing homodimer or heterodimer. The protein tyrosine kinase ZAP-70 binds to the phosphorylated immunoreceptor tyrosine-base activation motifs (ITAMs) of the TCR ζ chain through two Src-homology (SH2) domains. This binding results in the phosphorylation of ZAP-70 on multiple tyrosine residues, including Tyr 292 and Tyr 319. ZAP-70 is autophosphorylated on Tyr 292, which is thought to negatively regulate ZAP-70 function in lymphocytes. Alternatively, ZAP-70 is positively regulated by phosphorylation on Tyr 319, which mediates the SH2-dependent interaction between Lck and ZAP-70.

REFERENCES

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2. Frank, S.J., et al. 1990. The structure and signaling function of the invariant T cell receptor components. *Semin. Immunol.* 2: 89-97.
3. Watts, J.D., et al. 1994. Identification by electrospray ionization mass spectrometry of the site of tyrosine phosphorylation induced in activated Jurkat T cells on the protein tyrosine kinase ZAP-70. *J. Biol. Chem.* 269: 29520-29529.
4. Zhao, Q., et al. 1996. Enhancement of lymphocyte responsiveness by a gain-of-function mutation of ZAP-70. *Mol. Cell. Biol.* 16: 6765-6774.
5. Magistrelli, G., et al. 1999. Role of the Src homology 2 domains and interdomain regions in ZAP-70 phosphorylation and enzymatic activity. *Eur. J. Biochem.* 266: 1166-1173.
6. Di Bartolo, V., et al. 1999. Tyrosine 319, a newly identified phosphorylation site of ZAP-70, plays a critical role in T cell antigen receptor signaling. *J. Biol. Chem.* 274: 6285-6294.
7. Pelosi, M., et al. 1999. Tyrosine 319 in the interdomain B of ZAP-70 is a binding site for the Src homology 2 domain of Lck. *J. Biol. Chem.* 274: 14229-14237.

CHROMOSOMAL LOCATION

Genetic locus: ZAP70 (human) mapping to 2q11.2; Zap70 (mouse) mapping to 1 B.

SOURCE

p-ZAP-70 (Tyr 493) is a rabbit polyclonal antibody raised against a short amino acid sequence containing phosphorylated Tyr 493 of ZAP-70 of human origin.

PRODUCT

Each vial contains 100 μ g IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

APPLICATIONS

p-ZAP-70 (Tyr 493) is recommended for detection of Tyr 493 phosphorylated ZAP-70 of human origin, correspondingly phosphorylated Tyr 492 of mouse origin and correspondingly phosphorylated Tyr 488 of rat origin of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2 μ g per 100-500 μ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and immunohistochemistry (including paraffin-embedded sections) (starting dilution 1:50, dilution range 1:50-1:500); also recommended for detection of Tyr 526 phosphorylated Syk of human origin and correspondingly phosphorylated Tyr 520 of mouse and rat origin.

Suitable for use as control antibody for ZAP-70 siRNA (h): sc-29526, ZAP-70 siRNA (m): sc-36867, ZAP-70 shRNA Plasmid (h): sc-29526-SH, ZAP-70 shRNA Plasmid (m): sc-36867-SH, ZAP-70 shRNA (h) Lentiviral Particles: sc-29526-V and ZAP-70 shRNA (m) Lentiviral Particles: sc-36867-V.

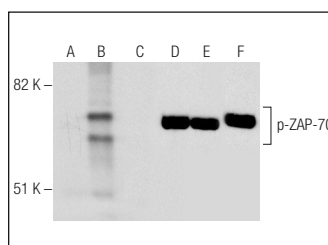
Molecular Weight of p-ZAP-70: 70 kDa.

Positive Controls: Jurkat whole cell lysate: sc-2204.

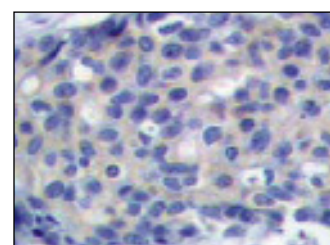
RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker[™] compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker[™] Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Western Blotting Luminol Reagent: sc-2048 and Lambda Phosphatase: sc-200312A. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz[™] Mounting Medium: sc-24941. 4) Immunohistochemistry: use ImmunoCruz[™]: sc-2051 or ABC: sc-2018 rabbit IgG Staining Systems.

DATA



Western blot analysis of ZAP-70 phosphorylation in untreated (A, D), pervanadate treated (B, E) and pervanadate and lambda protein phosphatase treated (C, F) Jurkat whole cell lysates. Antibodies tested include p-ZAP-70 (Tyr 493): sc-101823 (A, B, C) and ZAP-70 (1E7.2): sc-32760 (D, E, F).



p-ZAP-70 (Tyr 493): sc-101823. Immunoperoxidase staining of formalin-fixed, paraffin-embedded human breast carcinoma tissue showing cytoplasmic staining.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.