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Product Information

Anti-phospho-CD3ζ (pTyr¹⁴²) produced in rabbit, affinity isolated antibody

Catalog Number SAB4200334

Product Description

Anti-phospho-CD3 ζ (pTyr¹⁴²) is produced in rabbit using as immunogen a synthetic phosphopeptide corresponding to residues surrounding Tyr¹⁴² of mouse CD3 ζ (Gene ID: 12503) conjugated to KLH. The corresponding sequence is identical in human CD3 ζ . The antibody is affinity-purified using the immunizing peptide immobilized on agarose.

Anti-phospho-CD3 ζ (pTyr¹⁴²) recognizes mouse phosphorylated CD3 ζ (pTyr¹⁴²). The antibody may be used in several immunochemical techniques including immunoblotting and immunoprecipitation. Detection of the phospho-CD3 ζ (pTyr¹⁴²) by immunoblotting is specifically inhibited by the immunizing peptide and not inhibited by the corresponding non-phosphorylated peptide. In addition, binding is not inhibited by CD3 ζ -derived peptides that include phosphorylated Tyr⁷², Tyr⁸³, Tyr¹¹¹, Tyr¹²³ and Tyr¹⁵³.

Signal transduction by the T-cell antigen receptor (TCR) is initiated by a rapid and transient phosphorylation of tyrosine residues within conserved motifs (ITAMs) in the cytoplasmic tails of the CD3 subunits. These immunoreceptor tyrosine-based activation motifs (ITAMs) possess two repeats of the conserved sequence Tyr-X-X-Leu/lle spaced by 6-8 residues (where X is any amino acid). The phosphorylated ITAMs are critical for TCR signaling because they serve as temporary binding sites for cytoplasmic signaling effector molecules operating downstream of the activated receptor.¹²

The TCR-CD3 multimeric complex is composed of TCR $\alpha\beta$, CD3 $\gamma\epsilon$, CD3 $\delta\epsilon$ and $\zeta\zeta$ dimers. The CD3 γ , CD3 $\delta\epsilon$ and CD3 ϵ , each contain one ITAM, whereas the ζ chain contains three ITAMs. Amongst the TCR subunits, the CD3 ζ chain plays a key role in regulating TCR assembly, transport, and cell surface expression. Down-regulation of TCR ζ chain expression associated with T cell dysfunction was described in various pathologies.

Reagent

Supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide.

Antibody Concentration: ~ 1.0 mg/mL

Precautions and Disclaimer

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

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. 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

 $\label{eq:local_local_local_local} \underline{Immunoblotting} \hbox{: a working antibody concentration of } 0.5-1~\mu g/mL \hbox{ is recommended using lysates of } \\ HEK-293T \hbox{ cells co-expressing mCD3}\zeta \hbox{ and } \\ \text{constitutively active LCK.}$

Immunoprecipitation: a working amount of 1-2 μg is recommended using lysates of HEK-293T cells co-expressing mCD3 ζ and constitutively active LCK.

Note: In order to obtain the best results using various techniques and preparations and due to the high similarity in the amino acids sequence of the ITAMs we highly recommend determining the optimal working dilutions by titration.

References

- Underhill, D.M., and Goodridge, H.S., *Trends Immunol.*, 28, 66-73 (2007).
- 2. Love, P.E., and Hayes, S.M., Cold Spring Harb. Prespect. Biol., 2:a002485 (2010).
- 3. Baniyash, M., *Nat. Rev. Immunol.*, **9**, 675-687 (2004).

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