

Product Information

Anti-phospho-Integrin $\beta 1$ [pSer⁷⁸⁵]

Developed in Rabbit, Affinity Isolated Antibody

Product Number **I 7533**

Product Description

Anti-phospho-Integrin $\beta 1$ [pSer⁷⁸⁵] is developed in rabbit using a synthetic phosphorylated peptide derived from the region of human integrin $\beta 1$ containing serine 785 as immunogen. The antiserum is affinity purified using epitope-specific affinity chromatography. The antibody is preadsorbed to remove any reactivity toward a non-phosphorylated Integrin $\beta 1$.

The antibody detects chicken and mouse Integrin $\beta 1$. Human, rat and other species (100% homologous) of Integrin $\beta 1$ have not been tested, but are expected to react. Integrin $\beta 2$ (55%) and Integrin $\beta 7$ (64%) have also not been tested, but are not expected to react. The antibody has been used in immunoblotting applications.

Integrin $\beta 1$, also known as CD29, is a 130 kDa transmembrane glycoprotein that forms noncovalent complexes with various Integrin α subunits (including $\alpha 1$, $\alpha 2$, $\alpha 3$, $\alpha 4$, $\alpha 5$, and $\alpha 6$, also known as CD49a, CD49b, CD49c, CD49d, CD49e, and CD49f, respectively) to form the functional receptors that bind to specific extracellular matrix proteins. Integrin receptors are involved in the regulation of a variety of important biological functions, including embryonic development, wound repair, hemostasis, and prevention of programmed cell death. They are also implicated in abnormal pathological states such as tumor directed angiogenesis, tumor cell growth, and metastasis. These heterodimeric receptors bridge the cytoplasmic actin cytoskeleton with proteins present in the extracellular matrix and/or on adjacent cells. The clustering of integrins on a cell's surface leads to the formation of focal contacts. Interactions between integrins and the extracellular matrix lead to activation of signal transduction pathways and regulation of gene expression.

Phosphorylation of serine 785 on the Integrin $\beta 1$ promotes cell attachment, but inhibits spreading and migration, whereas dephosphorylation promotes cell spreading and migration.

Reagent

The antibody is supplied as a solution in Dulbecco's phosphate buffered saline (without Mg^{2+} and Ca^{2+}), pH 7.3, with 1.0 mg/ml BSA (IgG and protease free) and 0.05% sodium azide.

Precautions and Disclaimer

Due to the sodium azide content, a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazards and safe handling practices.

Storage/Stability

Store at $-70^{\circ}C$. Upon initial thawing freeze the solution in working aliquots for extended storage. Avoid repeated freezing and thawing to prevent denaturing the antibody. Do not store in frost-free freezers. Working dilution samples should be discarded if not used within 12 hours. The antibody is stable for at least 12 months when stored appropriately.

Product Profile

The supplied reagent is sufficient for 10 blots.

A recommended working concentration of 0.1 to 1.0 $\mu g/mL$ is determined by immunoblotting using fibroblasts lacking Integrin $\beta 1$ transfected with chicken Integrin $\beta 1$.

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working concentration by titration test.

Results

Peptide competition

1. Extracts prepared from Integrin $\beta 1$ -deficient fibroblasts expressing wild type chicken Integrin $\beta 1$ were resolved 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 follow:
 - Lane 1 no peptide
 - Lane 2 non phosphorylated peptide corresponding to the immunogen
 - Lane 3 a generic phosphoserine containing peptide
 - Lane 4 immunogen
4. All lanes were incubated with 0.50 µg/mL Integrin β1[pSer⁷⁸⁵] 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.

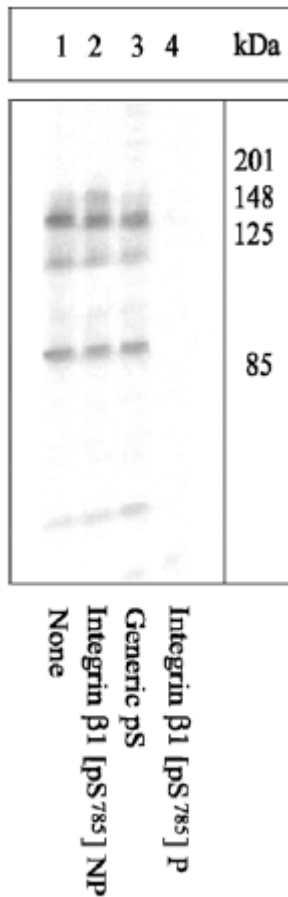


Figure 1 Peptide Competition

The data in Figure 1 show that only the peptide corresponding to Integrin β1[pSer⁷⁸⁵] blocks the antibody signal, thereby demonstrating the specificity of the antibody.

References

1. Mulrooney, J.P., et al. Serine 785 phosphorylation of the β1 cytoplasmic domain modulates β1A-integrin-dependent functions. *J. Cell Sci.*, **114**, 2525-2533 (2001).
2. Wennerberg, K., et al. Mutational analysis of the potential phosphorylation sites in the cytoplasmic domain of integrin β1A. *J. Cell Sci.*, **111**, 1117-1126 (1998)
3. Schlaepfer, D.D., et al. Signaling through focal adhesion kinase. *Prog. Biophys. Mol. Biol.* **71**, 435-478 (1999).

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