

Product Information

Anti-Phosphoserine Antibody, Mouse Monoclonal

Clone PSR-45, Purified from Hybridoma Cell Culture

P5747

Product Description

Anti-Phosphoserine Antibody, Mouse Monoclonal (IgG1 isotype) is derived from the PSR-45 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from a BALB/c mouse immunized with phosphoserine conjugated to KLH. The isotype is determined by a double diffusion assay using Mouse Monoclonal Antibody Isotyping Reagents, (ISO2).

Anti-Phosphoserine Antibody, Mouse Monoclonal recognizes phosphorylated serine both as a free amino acid or when conjugated to carriers such as BSA or KLH, using ELISA and dot blot. It does not react with non-phosphorylated serine, phosphorylated tyrosine or threonine, AMP, nor ATP. The antibody has been used for the detection of some phosphoserine containing proteins using immunoblotting^{1,2,8-10} and dot blotting.³

Protein phosphorylation and dephosphorylation are basic signaling mechanisms that modify protein function in eukaryotic cells.4 Phosphorylation is a rare post-translational event in normal tissues; however, the abundance of phosphorylated cellular proteins increases several fold following various activation processes. The main amino acids that are phosphorylated are tyrosine, serine, or threonine (pTyr/pSer/pThr), each having specific kinases that phosphorylate them and specific phosphatases that dephosphorylate them. Different growth factor receptors, such as the EGF-R, PDGF-R, and insulin receptor, contain Tyr/Ser/Thr residues that are autophosphorylated upon binding to their ligands.⁵ Ligand binding of other receptors, like the T cell antigen receptor complex or receptors for some hematopoietic growth factors, stimulate downstream associated kinases.6 An understanding of transformation by oncogenes and mitogenic processes of growth factors depends on the identification of their substrate and a subsequent determination of how phosphorylation affects the properties of these proteins.

Studies on the role of phosphorylated proteins have been hampered by their low abundance and the problem of distinguishing the various types of phosphorylated proteins. Immunoblotting of cellular proteins with antibodies directed against phosphoamino acids is advantageous over ³²P labeling and can therefore be employed to monitor alterations in phosphorylation of specific proteins as they occur in intact organs or even in whole animals. Indeed, mono- and polyclonal antibodies directed against phosphorylated residues were generated and found useful as analytical and preparative tools^{2,7} by enabling the identification, quantification, and immunoaffinity isolation of phosphorylated cellular proteins.

Reagent

Supplied as a solution in 0.01 M Phosphate buffered saline, pH 7.4, containing 15 mM Sodium azide as preservative.

Antibody Concentration

Approximately 2 mg/mL.

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

1

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 dilution samples should be discarded if not used within 12 hours.



Product Profile

Immunoblotting

A working concentration of 2.5–5 μ g/mL is determined using total rat brain extract.

Indirect ELISA

A working concentration of $0.3-0.6 \mu g/mL$ is determined using microwell plates coated with Phosphoserine-BSA, (P3717), $10 \mu g/mL$.

Note: In order to obtain best results in different techniques and preparations it is recommended to determine optimal working concentrations by titration test.

References

- Zacour, M.E. et al., Am. J. Physiol. Lung Cell. Mol. Physiol., 278, L59-L67 (2000).
- DeAizpurua, H.J. et al., J. Biol. Chem., 272, 16364-16373 (1997).
- 3. Pasqualini, E. et al., Biochem. J., 327, 527-535 (1997).
- 4. Hunter, T., and Cooper, J. A., Annu. Rev. Biochem., 54, 897-930 (1985).
- 5. Heffetz, D. et al., Methods Enzymol., 201, 44-53 (1991).
- 6. Alexander, D., and Cantrell, D., Immunol. Today, 10, 200-205 (1989).
- 7. Levine, L. et al., J. Immunol. Meth., 124, 239-249 (1989).
- 8. Lin, L. et al., Oncology Rep., 10, 665-669 (2003).
- 9. Zhao, M. et al., FEBS Lett., 509, 405-412 (2001).
- 10. Murphy, P.R. et al., Endocrinology, 142, 81-88 (2001).

Notice

We provide information and advice to our customers on application technologies and regulatory matters to the best of our knowledge and ability, but without obligation or liability. Existing laws and regulations are to be observed in all cases by our customers. This also applies in respect to any rights of third parties. Our information and advice do not relieve our customers of their own responsibility for checking the suitability of our products for the envisaged purpose.

The information in this document is subject to change without notice and should not be construed as a commitment by the manufacturing or selling entity, or an affiliate. We assume no responsibility for any errors that may appear in this document.

Technical Assistance

Visit the tech service page at SigmaAldrich.com/techservice.

Terms and Conditions of Sale

Warranty, use restrictions, and other conditions of sale may be found at <u>SigmaAldrich.com/terms</u>.

Contact Information

For the location of the office nearest you, go to SigmaAldrich.com/offices.

The life science business of Merck operates as MilliporeSigma in the U.S. and Canada.



