



Anti-Protein Phosphatase X/C Interior

Developed in Sheep, IgG Fraction of Antiserum

Product Number **P8859**

Product Description

Anti-Protein Phosphatase X/C Interior is developed in sheep using a highly purified peptide corresponding to amino acid residues 289-302 of protein phosphatase X/C as the immunogen.

Anti-Protein Phosphatase X/C Interior recognizes an internal sequence of protein phosphatase X catalytic domain (34 kDa) from human, mouse, rat and bovine by immunoblotting and immunocytochemistry.

The balance between protein kinase and phosphatase activities is responsible for controlling the level of protein phosphorylation and is a central mechanism controlling a wide range of cellular processes. Protein phosphatases are present in all eukaryotic cells and regulate several cellular processes among them cell-cycle progression, transcriptional regulation, cell growth, differentiation and apoptosis. The serine/threonine phosphatases have been classified into four groups which include PP1, PP2A, PP2B (also termed calcineurin) and PP2C on the basis of differences in their biochemical properties.^{1,2} Protein phosphatase 1, 2A and 2B are highly homologous members of the same family, but differ in their substrate specificity and interaction with regulatory molecules.^{2,3} PP2C appears to belong to an unrelated family.⁴

Protein Phosphatase 2A (PP2A) is a multimeric serine/threonine phosphatase that is implicated in numerous cellular processes including: cellular metabolism, DNA replication, transcription, RNA splicing, translation, cell-cycle progression, morphogenesis, development and transformation.^{5,6}

The PP2A holoenzyme consists of a catalytic subunit (C), a structural subunit (A) and a regulatory subunit (B). The diversity of PP2A substrates requires diversity of the enzyme. The C subunit of PP2A is well conserved, however. Instead, the B subunits regulate PP2A substrate specificity largely by targeting the holoenzyme to the proper cellular location.⁷

The cDNA for the catalytic domain PPX was first isolated from rabbit liver. PPX was found to be a protein serine/threonine phosphatase related to, but distinct from PP1 and PP2A.⁸ PPX is also called PP4 or PPP4.⁹ The complete amino acid sequence of PPX

Product Information

revealed that it is 49% identical to the catalytic subunit of PP1 and 69% identical to that of PP2A.^{8,10} The catalytic subunit of PPX and PP2A are identical at the extreme C-terminus.¹¹ The recombinant PPX catalytic subunit has substrate specificity and phosphatase inhibitor sensitivity similar to PP2A.¹⁰ Comparison of the amino acid sequences of the known phosphatases has resulted in the grouping of PPX into a subfamily with PP2A and another novel phosphatase PPV. The other subfamily composed of PP1 and the more recently identified PPY and PPZ.¹²

PPX/C is found dispersed throughout the cytoplasm and nucleus. It shows particularly intense staining at centrosomes, suggesting it may play a role in the control of microtubule nucleation.¹⁰ In support of this, a 50 kDa regulatory subunit of PPP4 (PPX) has been discovered that is localized at the centrosomes.⁹ Immunoblotting and immunocytochemistry indicate that PPX catalytic subunit is a predominantly nuclear phosphatase.¹³ This differs from PP2A which is dispersed throughout the cell, but located predominantly in the cytoplasm.¹⁴ Given this difference in subcellular distribution, it is possible that regulatory subunits and/or post-translational modifications aid in directing the catalytic subunits of these enzymes to specific regions in the cell. In fact, PPX, like PP2A, has been shown to be methylated at its C-terminus.¹⁵ Although the exact role of this modification is unknown, it is known for PP2A that the methylation state changes during the cell cycle.¹⁶ Furthermore, carboxymethylation has been shown to affect catalytic activity *in vivo*.¹¹

Reagents

Anti-Protein Phosphatase X/C Interior is supplied as 100 µg of sheep IgG at a concentration of 1 mg/ml in phosphate buffered saline with 0.08% 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 hazardous and safe handling practices.

Storage/Stability

Antibodies should be stored at –20°C. For extended storage, freeze in working aliquots. Repeated freezing and thawing is not recommended. 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

The recommended working dilution is 1:500 to 1:1000 for immunoblotting using peroxidase conjugated anti-sheep IgG and chemiluminescent detection. The recommended working dilution for immunocytochemistry is also 1:500 to 1:1000

Note: In order to obtain best results and assay sensitivities of different techniques and preparations, we recommend determining optimal working dilutions by titration test.

References

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