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Product Information

Monoclonal Anti-Protein Kinase C

Clone MC5

Mouse Ascites Fluid

Product Number **P 5704**

Product Description

Monoclonal Anti-Protein Kinase C (mouse IgG2a isotype) is derived from the MC5 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from an immunized mouse. Purified bovine brain protein kinase C was used as the immunogen.¹ The isotype is determined using Sigma ImmunoType™ Kit (Product Code ISO-1) and by a double diffusion assay using Mouse Monoclonal Antibody Isotyping Reagents (Product Code ISO-2).

Monoclonal Anti-Protein Kinase C (clone MC5) recognizes an epitope located within the amino acid sequence 296-317, at the hinge region, close to or at the trypsin cleavage site of protein kinase C (PKC).¹ The antibody reacts with the 80 kD polypeptide of PKC, applying the immunoblotting technique using bovine brain PKC, extracts of rat glioma and murine NIH 3T3 cell lines, and immunoprecipitation of rat glioma and human cell extracts (lysates). Binding of the antibody to purified PKC *in vitro*, blocks partial proteolysis by trypsin. Introduction of Fab fragment of the antibody into a rodent glioma cell line inhibits phorbol-ester-induced down-regulation of the kinase.¹ Cross-reactivity has been observed with human, bovine, rat and mouse. Also, the product may be used for immunohistochemical staining.

Monoclonal Anti-Protein Kinase C may be used for the localization of protein kinase C using various immunochemical assays such as ELISA, immunoblot, dot blot, immunoprecipitation, and immunohistochemistry.

Protein kinase C (PKC, 77-90 kDa) is one of a family of homologous serine-threonine protein kinases, that play a key role in signal transduction, cellular regulation, tumor promotion and oncogenesis.¹ PKC is a calcium-dependent and phospholipid-dependent enzyme that is activated *in vivo* by the lipid diacylglycerol, produced in response to a variety of hormones and growth factors.¹⁻³ PKC consists of a single polypeptide chain, containing four conserved regions and five variable regions. Sequence information defines a putative

domain structure for the enzyme which can be divided into an amino-terminal regulator and a carboxy-terminal catalytic domain joined by a hinge region. Proteolysis of purified native PKC by trypsin yields two major fragments, representing the regulatory and kinase domains of the enzyme, due to cleavage in the proposed hinge region between residue 292 and residue 317.⁴ There is evidence that *in vivo* agonist-induced generation of a catalytic fragment of the enzyme occurs as well.¹ The PKC family of isozymes can be subdivided into two major classes; conventional (C) isoforms (α , β_1 , β_2 , and γ), which are Ca^{2+} and phospholipid-dependent kinases, and novel (n) isoforms (δ , ϵ , ζ , η , and θ) that are Ca^{2+} -independent, phospholipid-stimulated kinases.⁵ Antibodies that react specifically with PKC are useful for the study of the specific activation requirements, subcellular distribution, substrate specificities, and variation in mode of action of these isoenzymes. They also allow the detection and localization of PKC in normal and malignant tissues.

Reagents

The product is provided as ascites fluid with 0.1% sodium azide as a preservative.

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

For continuous use, store at 2-8 °C for up to one month. For extended storage, solution may be frozen in working aliquots. Repeated freezing and thawing is not recommended. If slight turbidity occurs upon prolonged storage, clarify by centrifugation before use.

Product Profile

The minimum titer of 1:500 is determined by indirect immunoblotting using rat brain cytosol preparation.

In order to obtain optimum results it is recommended that each individual user determine their optimum working dilutions by titration assay.

References

1. Young, S., et al., Eur. J. Biochem., **173**, 247 (1988).
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3. Nishizuka, Y., Science, **233**, 305 (1986).
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5. Kiley, S. C., et al., J. Biol. Chem., **266**, 23761 (1991).

JWM/KMR 09/02

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