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ProductInformation

ANTI-CAM KINASE IV (CaMKIV), (AY-18)
Developed in Rabbit,
IgG Fraction of Antiserum

Product Number **C 2851**

Product Description

Anti-CaM Kinase IV (CaMKIV) (AY-18) is developed in rabbit using a synthetic peptide (K-AAVGLGVPPQQD-AILPEY) corresponding to the C-terminal region of rat CaM Kinase IV (amino acids 457-474 with N-terminal added lysine) conjugated to KLH as immunogen. This sequence has extensive homology (70%-80% identity) with human and mouse CaM Kinase IV. Whole antiserum is fractionated and then further purified by ion-exchange chromatography to provide the IgG fraction of antiserum that is essentially free of other rabbit serum proteins.

Anti-CaM Kinase IV (AY-18) recognizes rat CaMKIV by immunoblotting (65 kDa). The antibody cross-reacts with human CaMKIV. By immunoblotting, the CaMKIV band may appear as a doublet. Staining of CaMKIV in immunoblotting is specifically inhibited with CaMKIV immunizing peptide (CaMKIV, rat, amino acids 457-474 with N-terminally added lysine).

Anti-CaM Kinase IV (AY-18) may be used for the detection and localization of CAMKIV (65 kDa) by immunoblotting and immunohistochemistry.

Ca^{2+} /Calmodulin dependent protein kinases (CaM kinases, CaMKs) consist of a family of Ser/Thr protein kinases including CaMKI, CaMKII and CaMKIV.[1] CaMKs are considered to play a central role in many cellular functions in response to Ca^{2+} signaling, including synthesis and secretion of neurotransmitters, axonal transport, long term potentiation (LTP), receptor function, modification of the cytoskeleton and regulation of gene expression. CaM kinase IV (65 kDa) is abundantly expressed in the brain [2-5] (with the highest levels found in the cerebellum), in the thymus [6], and is localized to the cytoplasm and nucleus.[5,7] CaMKIV is thought to play a central role in controlling a wide range of Ca^{2+} mediated cellular functions in the central nervous system (CNS) and immune system. In the brain, it phosphorylates a wide range of substrates including synapsin I, microtubule associated protein (MAP2), tau and tyrosine hydroxylase. In Jurkat T lymphocytes, CaMKIV is activated through CD3-mediated signaling pathway.[8] CaMKIV is a monomeric enzyme containing a catalytic and autoinhibitory subunit. CaMKIV is transiently phosphorylated and activated, in response to rise in intracellular Ca^{2+}, by CaM kinase kinase (CaMKK), upstream in the CaMK signaling cascade.[8,9] CaMKIV translocates to the nucleus where it phosphorylates cAMP-response element-binding protein (CREB) at Ser^{133}, a key regulatory site controlling transcriptional activity.[10] CaMKIV phosphorylation and activity is regulated by the serine/threonine phosphatase PP2A, that dephosphorylates and inactivates CaMKIV. CaMKIV and PP2A have been shown to associate to form a stable signaling complex. [11]

Reagents

The product is supplied as IgG fraction in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM 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 hazardous 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 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

A minimum working dilution of 1:20,000 is determined by immunoblotting using a cytosolic fraction of rat brain extract.

A minimum working dilution of 1:1,000 is determined by immunoblotting using a whole cell extract of the human T-cell leukemia Jurkat cell line.

A minimum working dilution of 1:10,000 is determined by immunohistochemistry using formalin-fixed, paraffin-embedded sections of rat cerebellum.

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

References

1. Hanson, P.I. and Schulman, H., Ann. Rev. Biochem., **61**, 559 (1992).
2. Miyano, O., et al., J. Biol. Chem., **267**, 1198 (1992).
3. Okuno, S., et al., J. Biochem., **116**, 923 (1994).
4. Okuno, S., et al., J. Biochem., **119**, 1176 (1996).
5. Nakamura, Y., et al., Neuroscience, **68**, 181 (1995).
6. Frangakis, M.V., et al., J. Biol. Chem., **266**, 17592 (1991).
7. Jensen, K.F., et al., Proc. Natl. Acad. Sci. USA, **88** 2850 (1991).
8. Park, I-K, and Soderling, T.R. J. Biol. Chem., **270**, 30464 (1995).
9. Tokumitsu, H., et al., J. Biol. Chem., **270**, 19320 (1995).
10. Sheng, M., et al., Science, **252**, 1427 (1991).
11. Westphal, R.S., et al., Science, **280**, 1258 (1998).

lpg 9/99