CaM-kinase II (Human) Assay Kit

Cat. No. Kit-0146 **Lot. No.** (See product label)

SPECIFICATION

Product Overview	CaM-kinase II (Human) Assay Kit is a single-site, non-quantitative immunoassay for CaM kinase II activity. Plates are pre-coated with a newly designed Syntide-2, which can be efficiently phosphorylated by CaM kinase II on a microtiter plate. The detector antibody is MS-6E6, an antibody that specifically detects only the phosphorylated Syntide-2.
Description	Ca2+/CaM-dependent protein kinase II (CaM kinase II) is a ubiquitously expressed protein kinase that transduces elevated Ca2+ signals in cells to a number of target proteins ranging from ion channels to transcriptional activators. CaM kinase II has a unique holoenzyme structure and autoregulatory properties that allow it to give a prolonged response to transient Ca2+ signals and to sense cellular Ca2+ oscillations. In neurons CaM kinase II is highly expressed and localized with certain subcellular structures. Upon activation it can translocate to excitatory synapses where it regulates a number of proteins involved in synaptic transmission and its downstream signaling pathways. Changes in intracellular calcium can display variable responses ranging from highly localized, transient elevations within subcellular structures (e.g. a dendritic spine of a neuron) to Ca2+ waves that spread throughout the cell including the nucleus. The most ubiquitous calcium-sensing protein is Calmodulin (CaM), which contains four "EF" hand motifs with high specificity for binding Ca2+. The Ca2+/CaM complex interacts with and modulates the functionality of a large number of proteins including several Ser/Thr protein kinases.
Applications	1) Monitoring the purification of CaM kinase II.2) Screening inhibitors or activators of CaM kinase II3) Detecting the effects of pharmacological agents on CaM kinase II

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	activity.
Target Species	Human
Usage	For research use only (RUO)
Storage	• Upon receipt store the ATP at -20°C• Upon receipt store all other components at 4°C; Do not expose reagents to excessive light
Kit Components	Microplate: One microplate supplied ready to use, with 96 wells (12 strips of 8-wells) in a foil, zip-lock bag with a desiccant pack. Wells are coated with Syntide-2 as a substrate.10X Wash Buffer: One 100 mL bottle of 10X buffer containing 2%Tween -20Kinase Buffer: One bottle containing 20 mL of 1X buffer; used for Kinase Reaction Buffer and sample dilution.50X CaCl2: One vial containing 0.4 mL of 125 mM CaCl2, used for Kinase Reaction Buffer (Ca/CaM plus).50X EGTA: One vial containing 0.4 mL of 100 mM EGTA, used for Kinase Reaction Buffer (Ca/CaM minus).20X ATP: Lyophilized ATP Na2 salt. Reconstitute contents of vial with 1.6 mL of H2O. Mix gently until dissolved. Final concentration of ATP should be 1.25 mM ATP. The ATP solution can be stored in small aliquots (e.g. 100 µ) at -20°C. The 1.25 mM ATP stock solution must be diluted to 62.5 µM in Kinase Reaction Buffer (Ca/CaM plus) or Reaction Buffer (Ca/CaM minus) at the time of the assay. HRP conjugated Detection Antibody: One vial containing 12 mL of HRP (horseradish peroxidase) conjugated anti-phospho-Syntide-2 (MS-6E6) monoclonal antibody. Ready to use.Substrate Reagent: 20 mL of the chromogenic substrate, tetra-methylbenzidine (TMB). Ready to use.Stop Solution: One bottle supplied ready to use, containing 20 mL of 1 N H2SO4. Ready to use.

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