Casein Kinase II (CK2)

**Description:** Casein Kinase II (CK2) is a constitutively active serine/threonine protein kinase composed of two 44 kDa catalytic α-subunits and two 26 kDa regulatory β-subunits in an αβ configuration to form stable heterotetramers. CK2 holoenzyme undergoes autophosphorylation at two serine residues (S2/S3) of its β-subunit. Recently it has been shown that CK2 α-subunits undergo intermolecular tyrosine-autophosphorylation at Y182, which may represent a specific regulatory mechanism. Also, CK2 is able to phosphorylate, under special circumstances, tyrosyl residues in proteins. CK2 is implicated in a variety of cellular functions (1,2).

**Recognition Determinants:** The CK2 substrate specificity is invariably determined by multiple acidic residues located at positions between -2 and +5 relative to the target amino acid (mostly Ser and rarely Thr). The general recognition motif for phosphorylation by CK2 is SXXE/D, although SXE/D and S/D, and variations of these sequences are also phosphorylated. Polyanionic compounds, like heparin, inhibit CK2 activity with a Kᵣ of 1.4 nm (4,5).

**Source:** Isolated from a strain of *E. coli* expressing both α and β CK2 subunits derived from a human glioblastoma cDNA library (kindly provided by Dr. D. Marshak) (3).

Supplied in: 350 mM NaCl, 20 mM Tris-HCl (pH 7.5 @ 25°C), 1 mM Na₂EDTA, 2 mM DTT and 0.1% Triton X-100.

Reagents Supplied with Enzyme: 1X NEBuffer for Protein Kinases

**Reaction Conditions:** 1X NEBuffer for Protein Kinases, supplement with 200 μM ATP and gamma-labeled ATP to a final specific activity of 100–500 μCi/μmol. (CK2 will also accept GTP as a phosphoryl donor in place of ATP). **Incubate at 30°C.**

**Specific Activity:** ~ 589,000 units/mg.

**Molecular Weight:** α-subunit (45 kDa), β-subunit (25 kDa). The apparent molecular weight of the α-subunit estimated by SDS-PAGE is about 42 kDa.

**Quality Assurance:** CK2 contains no detectable protease or phosphatase activities.

**Quality Control Assays**

**Protease Activity:** After incubation of 5,000 units of Casein Kinase II (CK2) with 50 mM p-nitrophenyl phosphate for 2 hours at 30°C, no proteolytic activity could be detected by spectrophotometric analysis.

**Phosphatase Activity:** After incubation of 5,000 units of Casein Kinase II (CK2) with 50 mM p-nitrophenyl phosphate for 2 hours at 30°C, no phosphatase activity could be detected by spectrophotometric analysis.

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6. **Quality Control Assays**

   **Protease Activity:** After incubation of 5,000 units of Casein Kinase II (CK2) with a standard mixture of proteins for 2 hours at 30°C, no proteolytic activity could be detected by SDS-PAGE analysis.

   **Phosphatase Activity:** After incubation of 5,000 units of Casein Kinase II (CK2) with 50 mM p-nitrophenyl phosphate for 2 hours at 30°C, no phosphatase activity could be detected by spectrophotometric analysis.
References: