CREATIVE BIOMART®
Assay Kit

Polo-like kinase 1 (Human) Assay/Inhibitor Screening Assay Kit

Product Information

Cat.No. Kit-0705

Product Overview

Polo-like kinase 1 (Human) Assay/Inhibitor Screening Assay Kit is a single-site, non-quantitative immunoassay for Plk1 activity. Plates are pre-coated with a substrate corresponding to recombinant Protein-X, which contains threonine residues that can be efficiently phosphorylated by Plk1. The detector antibody specifically detects only the phosphorylated form of threonine residue on Protein-X.

Description

Polo-like kinases (Plk) have been shown to be important contributors to several cell-cycle events. Genetic and biochemical experiments in various organisms indicate that polo-like kinases regulate diverse cellular events at multiple mitotic stages. Genetic studies in Drosophila and yeast indicate plks function in centrosome assembly and separation during the formation of the bipolar spindle. Drosophila polo mutants reveal phenotypes of hyper-condensed chromosomes, monopolar spindles, disorganized spindle poles, and abnormal chromosome segregation. Schizosaccharomyces pombe plo1 displays similar phenotypes, such as the formation of monopolar spindles or a failure in septum formation after nuclear division. The budding yeast polo-like kinase homolog, Cdc5, seems to play an important role in actin ring formation and cytokinesis. In mammalian cells, antibody microinjection suggests a role for Plk1 in centrosome maturation. Mammalian Plk1 was further shown to phosphorylate specifically at least three components of APC, and to activate APC to ubiquitinate cyclin B in an in vitro-reconstituted system. More recent studies demonstrated that polo kinase activity plays a pivotal role in the separation of sister chromatids during mitosis.

Applications

1) Screening inhibitors or activators of Plk1.2) Detecting the effects of pharmacological agents on Plk1 activity.

Target Species

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Human

Usage

For research use only (RUO)

Storage

Upon receipt store the ATP at -20°CUpon 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 recombinant Protein-X as Plk1 substrate.10X Wash Buffer: One 100 mL bottle of 10X buffer containing 2%Tween -20.Kinase Buffer: One bottle containing 20 mL of 1X buffer; used for Kinase Reaction Buffer and sample dilution.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 µL) at -20°C. The 1.25 mM ATP stock solution must be diluted to 62.5 µM in Kinase Reaction Buffer at the time of the assay.Anti-Phospho-Threonine Polyclonal Antibody (PPT-07): One vial containing 12 mL of anti-phospho-threonine polyclonal antibody (PPT-07). Ready to use.HRP conjugated Anti-rabbit IgG: One vial containing 12 mL of HRP (horseradish peroxidase) conjugated anti-rabbit IgG. 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.

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