

p38 Kinase (Human) Assay/Inhibitor Screening Assay Kit

Cat. No. Kit-0495 **Lot. No.** (See product label)

SPECIFICATION

Product Overview

p38 Kinase (Human) Assay/Inhibitor Screening Assay Kit is a single-site, non-quantitative immunoassay for p38 activity. Plates are pre-coated with a substrate corresponding to recombinant ATF2, which contains threonine residues that can be efficiently phosphorylated by p38. The detector antibody specifically detects only the phosphorylated form of threonine residue on ATF2.

Description

So far, three distinct MAPK pathways have been described in mammalian cells: the extracellular signal-regulated kinases (ERKs) pathway, the c-Jun amino terminal kinase (JNK) pathway and the p38 MAPK pathway. In general, the ERKs are activated by mitogenic and proliferative stimuli, whereas the JNKs and p38 MAPKs respond to environmental stress, including ultraviolet light, heat, osmotic shock and inflammatory cytokines. Five homologues/isoforms of p38 MAPK have been identified: p38 α , p38 β , p38 γ , p38 δ and p38-2. The p38 MAPKs are activated by phosphorylation at the Thr-Gly-Tyr motif by the upstream kinases including MKK3 and 6. These upstream kinases have preferential effects on different p38 isoforms. Of these, p38 α is the best characterized and perhaps the most physiologically relevant kinase involved in inflammatory responses. The p38 α MAPK pathway is crucial to inflammatory cytokine production and signaling. Several p38 MAPK inhibitors have been shown to block the production of interleukin-1, tumour-necrosis factor and other cytokines. The inhibition of cytokine production seems to result from combined effects at the level of transcription and translation.

Applications

1) Screening inhibitors or activators of p38.2) Detecting the effects of pharmacological agents on p38 activity.

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Target Species	Human
Usage	For research use only (RUO)
Storage	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 recombinant ATF2 as p38 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 0.8 mL of H₂O. Mix gently until dissolved. Final concentration of ATP should be 2.5 mM ATP. The ATP solution can be stored in small aliquots (e.g. 100 µL) at -20°C. The 2.5 mM ATP stock solution must be diluted to 125 µM in Kinase Reaction Buffer at the time of the assay. Anti-Phospho-ATF2 Thr71 Polyclonal Antibody (PPT-09): One vial containing 12 mL of anti-phospho-ATF2 Thr71 polyclonal antibody (PPT-08). 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.