

Certificate of Analysis

CDK6/cyclin D3, 10 µg

Recombinant human full-length CDK6/cyclin D3



Part Number: A33517

Lot Number: 1861498

Immediate Storage: -80°C

Shipping Conditions: dry ice

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Description:

Recombinant human full-length active CDK6/cyclin D3 were co-expressed in insect cells using each with a N-terminal HIS tags.

CDK6 is a member of the cyclin-dependent family of protein kinases that are important regulators of cell cycle progression. CDK6 activity is regulated by the D-type cyclins and members of the INK4 family of CDK inhibitors. The CDK6 kinase activity is detected in mid-G1 phase of the cell cycle and is responsible for the phosphorylation and regulation of the activity of tumor suppressor protein Rb. Although CDK6 and CDK4 can both phosphorylate multiple residues in the Rb protein, they do so with different residue selectivities in vitro; CDK6 phosphorylates Thr821 while CDK4 phosphorylates Thr826 on Rb protein.

Accession Numbers:

The gene accession number for the subunits CDK4 and Cyclin D3 are NP_001250.1 and NP_001751.1.

Specific Activity:

40 nmoles of phosphate transferred to Rb (773-928) Recombinant Human C-terminal fragment per minute per mg of total protein at 30°C.

Concentration:

0.1 mg/mL total protein as measured using the Bradford protein assay with BSA as a standard.

Calculated **1,400 nM**.

Aliases:

CDK6: PLSTIRE, MGC59692

Cyclin D3: CCND3

Storage and Handling:

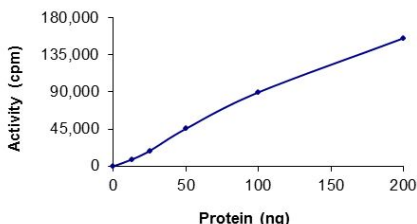
For maximum recovery please spin prior to use. Unless noted below, aliquots of the 5 µg, 10µg and 20µg sizes of kinase are not recommended as materials can be used in original packaging until exhausted. For larger sizes, the number of freeze/thaws may be reduced by preparing aliquots, aliquots below 20 µL are not recommended. **Please never store a kinase diluted.** If properly stored at -80°C, this product is guaranteed for 6 months from date of purchase.

Storage Buffer:

50 mM Tris-HCl (pH 7.5), 150 mM NaCl, 10 mM glutathione, 0.1 mM EDTA, 0.25 mM DTT, 0.1mM PMSF and 25% Glycerol.

QUALITY ASSURANCE

CDK6/cyclin D3 Activity Graph



Dilution Buffer:

5 mM MOPS (pH 7.2), 2.5 mM β-glycerol-phosphate, 5 mM MgCl₂, 1 mM EGTA, 0.4 mM EDTA and 50 ng/µL BSA.

Assay Conditions:

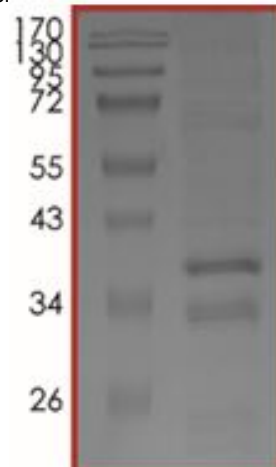
CDK6/cyclin D3 was pre-diluted in enzyme dilution buffer and assayed in 5 mM MOPS (pH 7.2), 2.5 mM β-glycerol-phosphate, 5 mM MgCl₂, 0.4 mM EDTA, 1 mM EGTA, 0.05 mM DTT, with 50 µM ATP, trace [³³P]-γ-ATP and 200 µg/mL Rb (773-928) Recombinant Human C-terminal fragment for 15 minutes at 30°C.

Gel Information for CDK6/cyclin D3

Page Description: Run on an SDS-PAGE gel and stained with Coomassie®.

Lane 1: Molecular Weight markers as labeled.

Lane 2: CDK6/cyclin D3



Purity:

> 75% as determined by a Coomassie® blue stained SDS-PAGE gel.

Molecular Weight:

38.0 / 33.6 kDa. Calculated from the protein sequence(s).

For questions, please contact our Technical Support Team

N. Am Ph#: 800-955-6288 or INTL Ph#: 760-603-7200 Select option 5, ext. 40266 Email: drugdiscoverytech@lifetech.com

Protein sequence alignment with reference sequence(s)

GenBank Accession Number: NP_001250.1

CDK6

1 MHHHHHSGMEKGLCRADQQYECVAEIGEGAYGKVFKARDLKNNGRFVALKRVVQTGEEGMPSTIREVAVLRHLETFEHPNVVRLFDVCTVSRTDRE Life CDK6/cyclin D3
1 M-----EKDGLCRADQQYECVAEIGEGAYGKVFKARDLKNNGRFVALKRVVQTGEEGMPSTIREVAVLRHLETFEHPNVVRLFDVCTVSRTDRE NP_001250.1
101 TKLTLVFEHVDQDLTTYLDKVPPEPGVPTETIKDMMFQLRGLDFLHSHRVVHRDLKPNILVTSSGQIKLADFLGARIYSFQMALTSVVVTLWYRAPEVL
92 TKLTLVFEHVDQDLTTYLDKVPPEPGVPTETIKDMMFQLRGLDFLHSHRVVHRDLKPNILVTSSGQIKLADFLGARIYSFQMALTSVVVTLWYRAPEVL
201 LQSSYATPVDLWSVGCIFAEMFRRKPLFRGSSDQVQKILQVIGLPGEDWPRDVALPRQAFHKSQAQIEKFTVDIDELGKDLLKCLTFNPAKRISA
192 LQSSYATPVDLWSVGCIFAEMFRRKPLFRGSSDQVQKILQVIGLPGEDWPRDVALPRQAFHKSQAQIEKFTVDIDELGKDLLKCLTFNPAKRISA
301 YSALSHPYFQDLERCKENLDSHLPSPQNTSELNTA
292 YSALSHPYFQDLERCKENLDSHLPSPQNTSELNTA

GenBank Accession Number: NP_00175.1

Cyclin D3

1 MHHHHHSGMEKGLCRADQQYECVAEIGEGAYGKVFKARDLKNNGRFVALKRVVQTGEEGMPSTIREVAVLRHLETFEHPNVVRLFDVCTVSRTDRE Life CDK6/cyclin D3
1 -----MELLCCGTRHAPRAGPDPRLLDGQVQLQSLRLLEERYVPRASYFQCVQREIKPHMRKMLAYWMLVCEEEQRCEEEVFPLAMNYLDRYLSC NP_00175.1
101 VPTRKAQLQLLGAVMMLASKLRETTPLTIEKLCIYTDHAVSPRQLRDWEVLVGLKQWDLAAVIAHDFLAFILHRLSLPRDRQALVKKHAQTFLALCAT
92 VPTRKAQLQLLGAVMMLASKLRETTPLTIEKLCIYTDHAVSPRQLRDWEVLVGLKQWDLAAVIAHDFLAFILHRLSLPRDRQALVKKHAQTFLALCAT
201 DYTFAMYPPSMIATGSIIGAAVQGLGACSMGDELTELLAGITGTEVDCLRACQEIEAALRESLREASQTSSSPAPKAPRGSSSQGPSQTSTPTDVTAIH
192 DYTFAMYPPSMIATGSIIGAAVQGLGACSMGDELTELLAGITGTEVDCLRACQEIEAALRESLREASQTSSSPAPKAPRGSSSQGPSQTSTPTDVTAIH
301 L
292 L

* highlighted residues denote differences from the reference protein sequence(s).

Anita Targosz

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Date: 16/Jan/2017

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For questions, please contact our Technical Support Team

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