## New England Biolabs Certificate of Analysis

| Product Name: | T4 Polynucleotide Kinase |
| :---: | :---: |
| Catalog Number: | M0201L |
| Concentration: | 10,000 U/ml |
| Unit Definition: | One Richardson unit is defined as the amount of enzyme catalyzing the incorporation of 1 nmol of acid insoluble $\left[{ }^{[33} \mathrm{P}\right]$ in 30 minutes at $37^{\circ} \mathrm{C}$. |
| Lot Number: | 10028773 |
| Expiration Date: | 06/2020 |
| Storage Temperature: | $-20^{\circ} \mathrm{C}$ |
| Storage Conditions: | 10 mM Tris-HCI , 50 mM KCl , 1 mM DTT, 0.1 mM EDTA , $0.1 \mu \mathrm{M}$ ATP, 50 \% Glycerol, (pH 7.4 @ $25^{\circ} \mathrm{C}$ ) |
| Specification Version: | PS-M0201S/L v1.0 |

T4 Polynucleotide Kinase Component List

| NEB Part Number | Component Description | Lot Number | Individual QC Result |
| :--- | :--- | :--- | :---: |
| M0201LVIAL | T4 Polynucleotide Kinase | 10008061 | Pass |
| B0201SVIAL | T4 Polynucleotide Kinase Reaction Buffer | 10016553 | Pass |


| Assay Name/Specification | Lot \# 10028773 |
| :---: | :---: |
| DNase Activity (Labeled Oligo, 5' extension) <br> A $50 \mu \mathrm{l}$ reaction in CutSmarte8 Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 5 ' extension and a minimum of 50 units of T4 Polynucleotide Kinase incubated for 16 hours at $37^{\circ} \mathrm{C}$ yields $<5 \%$ degradation as determined by capillary electrophoresis. | Pass |
| Double Stranded DNase Activity (Labeled Oligo) <br> A $50 \mu \mathrm{l}$ reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a blunt end and a minimum of 50 units of T4 Polynucleotide Kinase incubated for 16 hours at $37^{\circ} \mathrm{C}$ yields $<5 \%$ degradation as determined by capillary electrophoresis. | Pass |
| Endonuclease Activity (Nicking) <br> A $50 \mu \mathrm{l}$ reaction in T4 Polynucleotide Kinase Reaction Buffer containing $1 \mu \mathrm{~g}$ of supercoiled PhiX174 DNA and a minimum of 100 units of T4 Polynucleotide Kinase incubated for 4 hours at $37^{\circ} \mathrm{C}$ results in $<10 \%$ conversion to the nicked form as determined by agarose gel electrophoresis. | Pass |


| Assay Name/Specification | Lot \# 10028773 |
| :---: | :---: |
| Exonuclease Activity (Radioactivity Release) <br> A $50 \mu \mathrm{l}$ reaction in T4 Polynucleotide Kinase Reaction Buffer containing $1 \mu \mathrm{~g}$ of a mixture of single and double-stranded [ ${ }^{3} \mathrm{H}$ ] E. coli DNA and a minimum of 100 units of T4 Polynucleotide Kinase incubated for 4 hours at $37^{\circ} \mathrm{C}$ releases $<0.1 \%$ of the total radioactivity. | Pass |
| Non-Specific DNase Activity (16 Hour) <br> A $50 \mu \mathrm{l}$ reaction in T4 Polynucleotide Kinase Reaction Buffer containing $1 \mu \mathrm{~g}$ of Lambda DNA and a minimum of 100 units of T4 Polynucleotide Kinase incubated for 16 hours at $37^{\circ} \mathrm{C}$ results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis. | Pass |
| Protein Purity Assay (SDS-PAGE) <br> T4 Polynucleotide Kinase is $\geq 95 \%$ pure as determined by SDS-PAGE analysis using Coomassie Blue detection. | Pass |
| qPCR DNA Contamination (E. coli Genomic) <br> A minimum of 10 units of T4 Polynucleotide Kinase is screened for the presence of E . coli genomic DNA using SYBR® Green qPCR with primers specific for the E. coli 16S rRNA locus. Results are quantified using a standard curve generated from purified E . coli genomic DNA. The measured level of E . coli genomic DNA contamination is $\leq 1 \mathrm{E}$. coli genome. | Pass |
| RNase Activity (Extended Digestion) <br> A $10 \mu \mathrm{l}$ reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of $1 \mu$ l of T4 Polynucleotide Kinase is incubated at $37^{\circ} \mathrm{C}$. After incubation for 16 hours, $>90 \%$ of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection. | Pass |
| Single Stranded DNase Activity (FAM-Labeled Oligo) <br> A $50 \mu \mathrm{l}$ reaction in CutSmart(8) Buffer containing a 20 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 50 units of T4 Polynucleotide Kinase incubated for 16 hours at $37^{\circ} \mathrm{C}$ yields $<5 \%$ degradation as determined by capillary electrophoresis. | Pass |
| DNase Activity (Labeled Oligo, 3' extension) <br> A $50 \mu \mathrm{l}$ reaction in CutSmart(®) Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 3 ' extension and a minimum of 50 units of T4 Polynucleotide Kinase incubated for 16 hours at $37^{\circ} \mathrm{C}$ yields $<5 \%$ degradation as determined by capillary electrophoresis. | Pass |

This product has been tested and shown to be in compliance with all specifications.

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be INSPIRED
drive DISCOVERY stay GENUINE


Production Scientist
29 Jun 2018

## michael Oonello

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    Packaging Quality Control Inspector 10 Dec 2018

