NEW ENGLAND BioLabs Inc.
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# New England Biolabs
# Certificate of Analysis

| | |
|---|---|
| *Product Name:* | *KpnI-HF®* |
| *Catalog Number:* | *R3142L* |
| *Concentration:* | *20,000 U/ml* |
| *Unit Definition:* | *One unit is defined as the amount of enzyme required to digest 1 µg of pXba DNA in 1 hour at 37°C in a total reaction volume of 50 µl.* |
| *Lot Number:* | *10013138* |
| *Expiration Date:* | *04/2020* |
| *Storage Temperature:* | *-20°C* |
| *Storage Conditions:* | *50 mM KCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 µg/ml BSA* |
| *Specification Version:* | *PS-R3142S/L v1.0* |

**KpnI-HF® Component List**

| NEB Part Number | Component Description | Lot Number | Individual QC Result |
|---|---|---|---|
| R3142LVIAL | KpnI-HF® | 0091804 | **Pass** |
| B7204SVIAL | CutSmart® Buffer | 3081804 | **Pass** |
| B7024SVIAL | Gel Loading Dye, Purple (6X) | 0251805 | **Pass** |

| Assay Name/Specification | Lot # 10013138 |
|---|---|
| **Blue-White Screening (Terminal Integrity)**<br>A sample of Litmus28i vector linearized with a 10-fold excess of KpnI-HF™, religated and transformed into an E. coli strain expressing the LacZ beta fragment gene results in <1% white colonies. | **Pass** |
| **Endonuclease Activity (Nicking)**<br>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 100 Units of KpnI-HF™ incubated for 4 hours at 37ºC results in <10% conversion to the nicked form as determined by agarose gel electrophoresis. | **Pass** |
| **Exonuclease Activity (Radioactivity Release)**<br>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [ $^3$H] E. coli DNA and a minimum of 200 units of KpnI-HF™ incubated for 4 hours at 37ºC releases <0.1% of the total radioactivity. | **Pass** |
| **Ligation and Recutting (Terminal Integrity)**<br>After a 50-fold over-digestion of pXba DNA with KpnI-HF™, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16ºC. Of these ligated fragments, | **Pass** |

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| Assay Name/Specification | Lot # 10013138 |
|---|---|
| >95% can be recut with KpnI-HF™. | |
| **Non-Specific DNase Activity (16 Hour)**<br>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of pXba DNA and a minimum of 100 Units of KpnI-HF™ incubated for 16 hours at 37ºC results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis. | **Pass** |
| **Protein Purity Assay (SDS-PAGE)**<br>KpnI-HF™ is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection. | **Pass** |

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

Penghua Zhang
Production Scientist
18 Jun 2018

Mary Conlon
Packaging Quality Control Inspector
18 Jun 2018