

## New England Biolabs Certificate of Analysis

**Product Name:** *HphI*  
**Catalog Number:** *R0158S*  
**Concentration:** *5,000 U/ml*  
**Unit Definition:** *One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA in 1 hour at 37°C in a total reaction volume of 50 µl.*  
**Packaging Lot Number:** *10100720*  
**Expiration Date:** *03/2023*  
**Storage Temperature:** *-20°C*  
**Storage Conditions:** *300 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 500 µg/ml BSA*  
**Specification Version:** *PS-R0158S/L v2.0*

| HphI Component List |                       |            |                      |
|---------------------|-----------------------|------------|----------------------|
| NEB Part Number     | Component Description | Lot Number | Individual QC Result |
| R0158SVIAL          | HphI                  | 10100718   | Pass                 |
| B7204SVIAL          | CutSmart® Buffer      | 10097266   | Pass                 |

| Assay Name/Specification   | Lot # 10100720 |
|--|----------------|
| <b>Non-Specific DNase Activity (16 Hour)</b><br>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 25 Units of HphI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis. | Pass           |
| <b>Exonuclease Activity (Radioactivity Release)</b><br>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> H] E. coli DNA and a minimum of 5 units of HphI incubated for 4 hours at 37°C releases <0.2% of the total radioactivity.     | Pass           |
| <b>Ligation and Recutting (Terminal Integrity)</b><br>After a 5-fold over-digestion of Lambda DNA with HphI, ~50% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, ~75% can be recut with HphI.   | Pass           |

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

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16 Mar 2021



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