

## New England Biolabs Certificate of Analysis

**Product Name:** Tsp45I  
**Catalog Number:** R0583L  
**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 65°C in a total reaction volume of 50 µl.  
**Lot Number:** 10044943  
**Expiration Date:** 05/2021  
**Storage Temperature:** -20°C  
**Storage Conditions:** 20 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 0.15% Triton X-100, 200 µg/ml BSA  
**Specification Version:** PS-R0583S/L v1.0

| Tsp45I Component List |                       |            |                      |
|-----------------------|-----------------------|------------|----------------------|
| NEB Part Number       | Component Description | Lot Number | Individual QC Result |
| R0583LVIAL            | Tsp45I                | 10044944   | Pass                 |
| B7204SVIAL            | CutSmart® Buffer      | 10042965   | Pass                 |

| Assay Name/Specification   | Lot # 10044943 |
|--|----------------|
| <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 15 units of Tsp45I incubated for 4 hours at 65°C releases <0.1% of the total radioactivity.    | Pass           |
| <b>Ligation and Recutting (Terminal Integrity)</b><br>After a 10-fold over-digestion of Lambda DNA with Tsp45I, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, >95% can be recut with Tsp45I.  | Pass           |
| <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 15 Units of Tsp45I incubated for 16 hours at 65°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis. | Pass           |

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

*Ben Penta*

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Ben Penta  
Production Scientist  
08 May 2019

*Jay Minichiello*

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Jay Minichiello  
Packaging Quality Control Inspector  
22 May 2019