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

| | |
|---|---|
| *Product Name:* | *Bacteroides Heparinase III* |
| *Catalog Number:* | *P0737L* |
| *Concentration:* | *700 U/ml* |
| *Unit Definition:* | *One unit is defined as the amount of enzyme that will liberate 1.0 µmol unsaturated oligosaccharides from heparan sulfate per minute at 30°C and pH 7.0 in a total reaction volume of 100 µl.* |
| *Lot Number:* | *10033023* |
| *Expiration Date:* | *01/2020* |
| *Storage Temperature:* | *-80°C* |
| *Storage Conditions:* | *100 mM NaCl, 20 mM Tris-HCl, 1 mM EDTA, 5 mM CaCl2, (pH 7.5 @ 25°C)* |
| *Specification Version:* | *PS-P0737S/L v1.0* |

| **Bacteroides Heparinase III Component List** | | | |
|---|---|---|---|
| **NEB Part Number** | **Component Description** | **Lot Number** | **Individual QC Result** |
| P0737LVIAL | Bacteroides Heparinase III | 10033024 | **Pass** |
| B0735SVIAL | Bacteroides Heparinase Reaction Buffer (10X) | 0071803 | **Pass** |

| **Assay Name/Specification** | **Lot # 10033023** |
|---|---|
| **Glycosidase Activity (β1-3 Galactosidase)**<br>A 10 µl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC) and 1 unit of Bacteroides Heparinase III incubated for 20 hours at 30ºC results in no detectable activity as determined by thin layer chromatography. | **Pass** |
| **Glycosidase Activity (β1-4 Galactosidase)**<br>A 10 µl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-Galactosidase substrate (Galβ1-4GlcNAcβ1-3Galβ1-4Glc -AMC) and 1 unit of Bacteroides Heparinase III incubated for 20 hours at 30ºC results in no detectable activity as determined by thin layer chromatography. | **Pass** |
| **Glycosidase Activity (β-N-Acetylgalactosaminidase)**<br>A 10 µl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-N-Acetylgalactosaminidase substrate (GalNAcβ1-4Galβ1-4Glc-AMC) and 1 unit of Bacteroides Heparinase III incubated for 20 hours at 30ºC results in no detectable activity as determined by thin layer chromatography. | **Pass** |

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| Assay Name/Specification | Lot # 10033023 |
|---|---|
| **Glycosidase Activity (β-N-Acetylglucosaminidase)**<br>A 10 µl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled β-N-Acetylglucosaminidase substrate (GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC) and 1 unit of Bacteroides Heparinase III incubated for 20 hours at 30ºC results in no detectable activity as determined by thin layer chromatography. | **Pass** |
| **Protease Activity (SDS-PAGE)**<br>A 20 µl reaction in 1X Heparinase Reaction Buffer containing 24 µg of a standard mixture of proteins and a minimum of 5 units of Bacteroides Heparinase III incubated for 20 hours at 37°C, results in no detectable degradation of the protein mixture as determined by SDS-PAGE with Coomassie Blue detection. | **Pass** |
| **Protein Purity Assay (SDS-PAGE)**<br>Bacteroides Heparinase III is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection. | **Pass** |
| **Sulfatase Activity (2-O)**<br>A 10 µl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled 2-O-Sulfatase substrate (ΔUA2S-(1-4)-GlcNS6S-AMC) and 1 unit of Bacteroides Heparinase III incubated for 20 hours at 30ºC results in no detectable activity as determined by thin layer chromatography. | **Pass** |
| **Sulfatase and Uronidase Activity (N,6-O)**<br>A 10 µl reaction in Heparinase Reaction Buffer containing 1 nM of fluorescently-labeled N,6-O-Sulfatase substrate (ΔUA-(1-4)-GlcNS6S-AMC) and 1 unit of Bacteroides Heparinase III incubated for 20 hours at 30ºC results in no detectable activity as determined by thin layer chromatography. | **Pass** |

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

Brad Landgraf
Production Scientist
09 Jan 2019

Michael Tonello
Packaging Quality Control Inspector
09 Jan 2019